MYCOLOGIA

OFFICIAL ORGAN OF THE MYCOLOGICAL SOCIETY OF AMERICA

Vol. LI JULY-AUGUST, 1959 No. 4

CONTENTS

The effects of various reagents, mounting media, and dyes on the teliospore walls of Tilletia contraversa Kühn
SHIRL O. GRAHAM 477

Filamentous fungi from blossoms, ovaries, and fruit of pickling cucumbers
F. L. RAYMOND, J. L. ETCHELLS, T. A. BELL, AND P. M. MASLEY 492

Some viewpoints on the phylogeny of rust fungi. III. Origin of grass rusts ..... E. E. LEPPIK 512

The agaric flora of Texas. III. New taxa of brown- and black-spored agarics ..... HARRY D. THIERS 529

Studies of some Tremellaceae. III. The genus Bourdotia
KENNETH WELLS 541

Notes on boletes. XI.
WALTER H. SNELL, ROLF SINGER, AND ESTHER A. DICK 564

New and interesting species of Basidiomycetes VII
ROLF SINGER 578

Notes and Brief Articles 595

van Uden and do Carmo-Sousa on *Geotrichum candidum*; Fitt on Myxomycetes; Baxter on rusts.

Reviews 602

[MYCOLOGIA for May-June (51: 311-476) was issued December 8, 1959]

PUBLISHED BIMONTHLY FOR
THE NEW YORK BOTANICAL GARDEN
AT PRINCE AND LEMON STS., LANCASTER, PA.

Second-class postage paid at Lancaster, Pa.

MYCOLOGIA

Published by

THE NEW YORK BOTANICAL GARDEN

IN COLLABORATION WITH THE

MYCOLOGICAL SOCIETY OF AMERICA

OFFICERS OF THE MYCOLOGICAL SOCIETY OF AMERICA

CONST. J. ALEXOPOULOS, *President*
State University of Iowa

LINDSAY S. OLIVE, *Vice-President*
Columbia University

JOHN R. RAPER, *Past President* (1958)
Harvard University

C. W. HESSELTINE, *Councilor*, 1958–59
Northern Regional Research and Development Div., U.S.D.A.

W. G. SOLHEIM, *Councilor*, 1959–60
University of Wyoming

CHESTER W. EMMONS, *President-elect*
National Institutes of Health

E. S. BENEKE, *Sec.-Treas.*
Michigan State University
East Lansing, Michigan

D. P. ROGERS, *Past President* (1957)
University of Illinois

HELEN M. GILKEY, *Councilor*, 1958–59
Oregon State College

EDITORIAL BOARD

DONALD P. ROGERS, *Editor-in-Chief*
Department of Botany
University of Illinois
Urbana, Illinois

CLARK T. ROGERSON, *Managing-Editor*
New York Botanical Garden
Bronx Park, New York 58, N. Y.

ALMA WHIFFEN BARKDALE, '59
New York Botanical Garden,
Bronx Park, New York 58, N. Y.

EDITH K. CASH, '60
Plant Industry Station, Beltsville, Maryland

C. W. EMMONS, '61
National Institutes of Health
Bethesda 14, Maryland

ROY F. CAIN, '62
University of Toronto
Toronto 5, Canada

H. L. BARNETT, '63
West Virginia University, Morgantown, W. Va.

MYCOLOGIA

OFFICIAL ORGAN OF THE MYCOLOGICAL SOCIETY OF AMERICA

VOL. LI JULY–AUGUST, 1959 No. 4

THE EFFECTS OF VARIOUS REAGENTS, MOUNTING MEDIA, AND DYES ON THE TELIOSPORE WALLS OF TILLETIA CONTRAVERSA KÜHN [1]

SHIRL O. GRAHAM

(WITH 1 FIGURE)

INTRODUCTION

In recent years the structure and composition of the smut teliospore wall has received scant attention. Components of these walls such as reticulations and sheaths, including their configurations, are recognized by mycologists as critical taxonomic characters in many genera including *Tilletia*. However, very little is known about their responses to some of the standard micro-technical reagents. Consequently, the appearance of the teliospore wall after treatment with various reagents may be confusing in morphologic determinations or comparisons.

One has but to compare spores of dwarf bunt, *Tilletia contraversa* Kühn, from a given sorus mounted in lactophenol, having a pH of about 2, with spores from the same sorus mounted in 5% potassium hydroxide, having a pH of about 13, to be aware of the effects of pH on sheath and reticular contour. The taxonomic significance is obvious, yet little attention has been given to such a simple factor as the pH of mounting medium.

[1] Scientific Paper No. 1844 Washington Agricultural Experiment Stations, Project 1285.

[MYCOLOGIA for May–June (51: 311–476) was issued December 8, 1959]

In taxonomic studies of smuts, hydrous mounting media are a must, since morphological characters are distorted or completely contracted in anhydrous reagents. This suggests a direct correlation between spore-wall expansion and the degree of hydration allowed by any given mounting medium. Furthermore, variable pH effects suggest differential expansions among spore-wall components in the diverse chemicals and solvents currently used to formulate hydrous mounting media.

If morphological contour is altered by variables inherent in diverse mounting media, it is apparent that precise conditions and procedures should be set forth so as to reduce confusion. Otherwise, the validity of taxonomic concepts based on observed differences in sheath and wall configurations is questionable. The intent of this study was to evaluate some of the more common reagents used in accepted mounting media, to compare the currently accepted mounting media, and to suggest certain standardizations. Also, as an aid in defining the characters of the sheath and the areolae, an appropriate staining schedule which would not produce artifacts was sought. The use of aqueous dyes to accentuate spore-wall parts has numerous disadvantages. Often the uptake of such dyes by different components of the spores is erratic, and affinities of the various wall parts for specific dyes usually overlap. As an additional complicating factor, counterstaining is limited, owing to the incompatibilities of many aqueous dyes, and also to pH relationships.

Since the dwarf bunt fungus, *Tilletia contraversa* Kühn, has a well-sculptured reticulum and also possesses a hygroscopic sheath, it was selected as an appropriate test organism for these studies.

MATERIALS AND METHODS

The teliospore materials used in these studies were obtained from various field collections [2] of dwarf bunt. Only well-formed and apparently well-matured sori were selected unless otherwise indicated.

In evaluating the effect of pH, obviously neutrality is the base from which variability attributable to pH should be measured. Consequently, the mean spore-wall thickness was determined from the measurements of 100 teliospores mounted and warmed to boiling in pH 7 McIlvaine's buffer solution. This buffer was selected since there was no apparent difference between mean wall thicknesses whether it or doubly distilled water of pH 6.7 was used.

Preliminary studies showed that a general mean for sheath and reticular thicknesses could not be established for this species. For ex-

[2] Material collected and identified by the USDA Northwest Regional Smut Research Laboratory, Pullman, Washington.

ample, thicknesses were generally consistent for spores from a given sorus, but great variations occurred among sori of different collections. This was true even among individual sori from a single field collection. The relative maturity of sori apparently accounted in part for this variation. Mature sori in general contained spores with thinner sheaths and more shallow reticulations than did immature sori. Hence, the degree of expansion attributable to pH and other factors was considered relative only if measurements were made on spores from a single sorus. All measurements are presented on this basis.

The semi-permanent mounting media normally used in smut taxonomy are Shear's mounting fluid[3] (**4**), lactophenol (**5**, **7**), Karo corn syrup (**6**), and glycerine jelly (**6**). Temporary mounts are usually made in water or in 2.5–5% potassium hydroxide (**7**). It is standard practice to place smut spores in the given medium and then to warm the mount to its boiling point. The heating, in addition to eliminating air, is said to cause rapid expansion of the hygroscopic wall parts or to increase definition of them if the medium has that capacity. No reference study on this procedure was found in the literature, and so it was evaluated in addition to the other observations. Thus, to make valid comparisons of sheath and reticular responses to various reagents, the following testing procedures were used:

Spores from a single sorus were placed on a slide, immersed in two drops of the freshly prepared reagent to be tested, covered with a coverslip, allowed to stand for 15 minutes, observed, and measured at ×660 magnification with a filar micrometer. Then the slide was warmed to the boiling point of the reagent over an alcohol lamp; the slide was allowed to cool, and observations and measurements were again taken.

Measurements were made on 100 spores in each mount. Two measurements were made on each spore, thickness of the sheath, and the thickness of the reticulum layer. Since measurements were for relative comparisons from a given sorus, actual values in microns were not computed. A mean was established for each group of measurements in vernier units of the micrometer employed. This mean value was the datum unit

[3] Shear's mounting fluid described by Chupp (**1**) is only slightly different from what Diehl (**3**) used for semipermanent slide mounts as "Patterson's mounting medium." Chupp's formula is used at this institution.

Chupp's Formula		*Diehl's Formula*	
2% potassium acetate (in water)	300 ml	Potassium acetate	10 gm
Glycerine	120 ml	Water	500 ml
95% ethyl alcohol	180 ml	Glycerine	200 ml
		95% ethyl alcohol	300 ml

selected for appraising spore-wall response to the various reagents and was rounded off to the closest digit.

THE RESPONSE OF WALL PARTS TO REAGENTS

Before comparing the mounting media in common use, the physical responses of sheath and reticular materials to various reagents incorporated into such mounting media needed appraisal. It was considered that this would pinpoint any reagents that might mask, accentuate or counteract the individual effects of other reagents in complex formulations.

The reagents were prepared in the concentrations commonly used in the various mounting media. In addition, pH 7 McIlvaine's buffer solution, selected acids, bases and oxidizing agents were included to allow observation of wall-part reaction to differing pH values and to differing ions. Materials used were CP grade or redistilled. Doubly distilled water was used as a solvent or diluent. The pH of a freshly prepared reagent and the pH of the same reagent after rapidly heating it to boiling were determined.

The reagents tested and the results of this study are summarized in TABLE 1. From these measurements and observations it is not certain that pH was a contributing variable. The primary factor determining sheath expansion apparently is its ability to absorb water from the medium in which it is placed. The swelling effects of alkalies, the attenuating effects of acids and the responses of the material to oxidizing agents depended upon the reagent used. Apparently glycerine would substitute for water, at least in part, after heating. The obvious dehydrating effect of ethyl alcohol is clear. Greater expansion of sheath material after heating in this reagent can be attributed to a reduction in its concentration.

Reticular materials responded independently to the reagents tested, in contrast with sheath material, indicating a different chemical context. Reticular materials did not appear to be greatly altered by acids, although most bases and oxidizing agents distended the outer periphery of the structure more. They also outlined areolar topography exceedingly well.

It is also apparent that warming spores in most reagents affected expansion. With all reagents except potassium chloride and one per cent phenol, heating increased the overall expansion. It is presumed that shrinkage after heating in these cases was caused by an independent response of a wall structure to the concentration of the reagent concerned. The presumption is further attested by an apparent shrinkage

of sheath material after heating, although the overall expansion of the two components together was greater.

Reagents differed in their abilities to clear teliospores for observations of fine detail. The last two columns of TABLE 1 are based on arbitrary values for this capacity. Spores mounted in cold, doubly distilled water and subsequently warmed to boiling were the base from which comparisons were made. The clarity of such a warmed mount

TABLE 1

THE EXPANSION AND CLARITY OF TELIOSPORE-WALL PARTS OF TILLETIA CONTRAVERSA IN VARIOUS REAGENTS AND THE HYDROGEN-ION CONCENTRATION OF THE REAGENTS BEFORE AND AFTER HEATING TO BOILING

Reagent	Cold			Heated to boiling			Relative clarity	
	Sheath*	Reticulum	pH	Sheath	Reticulum	pH	Before heat	After heat
Distilled water (doubly)	30	39	6.4	30	40	6.8	−	0
McIlvaine's buffer (pH 7)	30	39	7.0	29	40	7.0	0	+
Potassium chloride (0.2/M)	25	34	7.1	25	31	6.9	0	0
Potassium hydroxide (0.2/M)	30	40	13.0	32	41	13.6	++	+++
Ammonium hydroxide (4%)	31	39	11.7	32	41	11.2	++	+++
Sodium bicarbonate (0.2/M)	32	36	10.0	31	39	10.1	++	++
Potassium acetate (0.3/M)	29	40	7.3	29	41	7.6	0	++
Potassium permanganate (1%)	30	35	9.3	30	42	9.1	− −	−
Sodium hypochlorite (1%)	31	40	11.7	30	43	10.1	+++	+++
Phenol (1%)	25	39	7.4	21	40	6.4	0	0
Phenol (20%)	18	35	4.6	18	40	4.5	0	0
Acetic acid (5%)	22	38	2.6	27	39	2.4	−	+
Lactic acid (20%)	12	37	1.7	18	39	1.6	+	++
Glycerine (10%)	29	39	6.5	27	41	5.4	+	++
Glycerine (20%)	29	38	6.0	27	41	5.2	++	+++
Glycerine (40%)	28	36	6.4	26	40	4.8	+	+++
Glycerine (50%)	29	34	5.9	24	41	4.6	+	+++
Glycerine (70%)	14	35	5.7	24	40	4.6	−	+++
Glycerine (100%)	immeasurable		4.6	20	39	4.6	− − −	+++
Alcohol (10%)	21	38	7.0	30	38	7.1	−	0
Alcohol (20%)	13	34	7.1	27	38	7.0	−	0
Alcohol (30%)	13	34	7.2	24	37	7.0	−	0
Alcohol (40%)	12	32	7.1	18	35	7.0	− −	−
Alcohol (50%)	9	32	6.9	14	34	7.0	− − −	− −
Alcohol (70%)	immeasurable		7.0	7	32	7.0	− − −	− − −
Alcohol (80%)	immeasurable		7.0	immeasurable		7.0	− − −	− − −
Alcohol (95%)	immeasurable		7.0	immeasurable		7.0	− − −	− − −

* Mean values for 100 spores in vernier units of a filar micrometer, magnification ×660. Entries marked immeasurable were so owing to inability to differentiate sheath and reticular components.

was given the arbitrary rating of zero. All other reagents were compared with this standard and given a relative clarity as indicated in the table, plus values for more clarity and minus values for less clarity. The data show that warming spores in most of the reagents tested enhanced spore definition. In no case did warming reduce clarity of the mount.

From this study it is concluded that any mounting medium used in smut taxonomy should be one that does not curtail nor exaggerate expansion of wall parts; it should contain a clearing agent; and, spores should be warmed in the medium before measurements are taken.

THE RESPONSE OF WALL PARTS TO CURRENTLY ACCEPTED SEMIPERMANENT MOUNTING MEDIA

A. Responses to Buffering

The possible contributing variables of pH and the synergistic effects of two or more reagents as they are combined in any given mounting medium needed clarification. A study was made comparing the variously used semipermanent mounting media (Shear's mounting fluid, lactophenol, glycerine jelly, and Karo corn syrup). These media were prepared according to formula and compared with the same media prepared with pH 7 McIlvaine's buffer in place of the water fraction.

TABLE 2

THE RELATIVE EXPANSIONS OF TELIOSPORE-WALL PARTS IN VARIOUS MOUNTING MEDIA AND THE INEFFECTIVENESS OF BUFFERING THE MEDIUM

Mounting medium	Cold			Heated		
	Sheath[a]	Reticulum	pH	Sheath	Reticulum	pH
McIlvaine's Buffer (pH 7)	30	39	7.0	29	40	7.0
Karo Corn Syrup (15%)	30	36	4.8	30[b]	37	4.4
Karo Corn Syrup (15%) plus Buffer	27	38	6.8	30[b]	40	7.0
Glycerine Jelly	—	—	6.2	29	38	5.2
Glycerine Jelly plus Buffer	—	—	6.9	29	39	5.2
Lactophenol	18	38	1.9	20	40	2.6
Lactophenol plus Buffer	22	39	4.1	21	40	2.6
Shear's Mounting Fluid	27	39	6.4	30	41	8.2
Shear's Mounting Fluid plus Buffer	30	38	7.2	30	41	8.2

[a] Mean values for 100 spores in vernier units of a filar micrometer rounded off to nearest digit, mag. ×660.

[b] Spores collapsed on one side owing to inability of material to penetrate into center of spore.

Mounts were made of teliospores from a single mature sorus in the same manner as described above for reagents. All mounting media were freshly prepared at the start of the study. The sorus selected for the study was one whose average spore measurements in the buffer were comparable with those from the sorus used in the previous study. The thicknesses of the sheath and of the reticulum were measured. An average was obtained according to the method outlined earlier, both for spores mounted in the cold medium and again after heating the mount to boiling. An aliquot of each mounting medium was measured for pH, both before and after heating to 100° C. The results of the study are presented in TABLE 2.

From the standpoint of clarity of the mount, preference was in the following descending order: 1) Shear's mounting fluid, 2) lactophenol,

3) glycerine jelly, 4) Karo corn syrup, and 5) pH 7 McIlvaine's buffer. The buffer seemed to have no effect on clarity when incorporated into any of the media. Karo corn syrup is not a suitable medium because spores collapse when they are heated in it. Apparently this is owing to the inability of the medium to infiltrate rapidly enough into the central portions of the spores. Once the spores had collapsed they could not be expanded again. Shear's mounting fluid not only cleared spores much better than any other mounting medium, but it also was superior for distinguishing areolar topography and reticular contour.

Preparing any given medium tested with 0.2 M McIlvaine's pH 7 buffer had no ultimate effect except on corn syrup. Here the hydrogen-ion concentration remained at pH 7 after heating, and the indications were that pH might be a contributing factor if the hydrogen-ion concentration could be stabilized.

B. An Evaluation of Shear's Mounting Fluid

Of all media tested, Shear's mounting fluid gave the best definition of spore wall sculpturing; therefore, this medium was considered in detail. Factors needing clarification were: 1) the independence of pH as a contributing variable, 2) the heating as an independent variable, and 3) the probability of an expansion sequence in a medium containing alcohol, water and glycerine. The last variable involves synergistic possibilities as well as the degree of constituent volatilization when heated to boiling.

The pH of freshly prepared medium, of one-year-old media stored in glass in both the light and in darkness, of two-year-old medium stored in glass in the dark, and of ingredient combinations of the medium were determined before and after heating to remove first the alcohol (100°) and then the water (130°). These readings are shown in TABLE 3.

Upon removal of alcohol from Shear's mounting fluids tested, the pH was near neutrality, but on continued heating to remove water, the pH always raised to pH 8.2, as it had in all previous studies. The combination of glycerine and potassium acetate apparently caused this result. In any event, it is important to point out the consistent pH of 8.2 as the end result when both alcohol and water were removed. Consequently, it is plausible that if some water was not removed, the pH would tend to be more neutral, a possible contributing variable attributable to pH.

If pH would strongly affect wall expansion, or if heating to remove all the water and alcohol from a mount would cause shrinkage of already expanded parts, a series of differentially buffered media should disclose

this state in the water-glycerine phase after alcohol removal but before the water fraction had been driven off. Hence, a series of Shear's mounting fluids was made up in which 0.2 M buffer solutions,[4] ranging from pH 4 to pH 10, were substituted for the water portion of the formulation.

TABLE 3

THE pH OF SHEAR'S MOUNTING FLUID AND ITS COMPONENTS IN RELATION TO REMOVAL OF SOLVENTS BY HEATING

Ingredient combination or formulation	Initial pH	pH after heating to remove alcohol (100° C)	pH after heating to remove water (130° C)
Freshly prepared medium	6.4	6.9	8.2
One-year-old medium, glass-stoppered, held in dark	6.7	6.8	8.2
One-year-old medium, glass-stoppered, held in light	6.5	6.8	8.2
Two-year-old medium, glass-stoppered, held in dark	6.8	7.1	8.2
Two % potassium acetate	7.8	—	—
Two % potassium acetate plus ethyl alcohol	7.3	7.7	—
Ethyl alcohol	7.0	—	—
Glycerine plus two % potassium acetate	6.1	—	8.2
Glycerine plus alcohol	5.2	4.6	—
Glycerine (C. P. grade)	4.6	—	4.6

Spores from a single sorus were mounted in each mixture of the series and the means were established for 100 spores as previously indicated. Each mount in turn was then placed on a warming plate, gradually heated to 90°, and then rapidly cooled by transfer onto an inverted tray of ice cubes. It was assumed that this would prevent excessive water loss of the mounts. Measurements were taken again and each mount in turn was heated to 130° on the warming plate to eliminate water. Measurements were taken again. Concurrently, the pH of each buffered solution was measured before heating, after heating to drive off the alcohol, and after heating to drive off the water.

The results (TABLE 4) show that pH does affect the amount of expansion of spore wall materials. In performing the mechanics of producing semipermanent spore mounts using Shear's medium an error may be injected unless all the alcohol and water are removed. If they are

[4] Prepared from pHydrion Buffer Powders obtained from Micro Essential Laboratories, Brooklyn 10, N. Y. It is realized that possible contributing variables have been introduced that are indigenous to the materials involved, especially in making up the higher pH formulations of the series. However, for this study it was not deemed too significant a factor.

not eliminated, the pH will be less than 8.2, resulting in less expansion of the sheath component in particular. The data show that a buffered medium of pH 8 before water removal is close to the end result of most of the series tested after alcohol and water extraction. Hence, it would seem advisable to buffer Shear's mounting fluid at pH 8 so that error

TABLE 4

TELIOSPORE-WALL EXPANSION IN SHEAR'S MOUNTING FLUID AS AFFECTED BY DIFFERENT HYDROGEN-ION CONCENTRATIONS

0.2/M Standard Buffer Series	Before heating			After heating to 90° C			After heating to 130° C		
	Sheath*	Reticulum	pH	Sheath	Reticulum	pH	Sheath	Reticulum	pH
Unbuffered	23	43	6.4	27	45	7.3	30	45	8.2
pH 4	26	38	4.7	26	42	4.3	30	44	8.1
pH 5	27	41	5.1	25	43	5.0	29	46	8.1
pH 6	27	40	5.4	27	44	6.0	30	45	8.2
pH 7	25	42	6.7	28	44	6.8	30	45	8.3
pH 8	25	44	7.6	30	45	7.9	29	46	8.3
pH 9	25	45	8.4	29	47	9.0	30	48	9.2
pH 10	24	45	8.9	30	47	10.1	30	48	10.2

* Mean values for 100 spores in vernier units of a filar micrometer rounded off to nearest digit, mag. ×660.

would be avoided should some water not be eliminated in mount preparation. Just why the pH rose after alcohol and water removal is not understood. The rise probably is due to dissociation of the potassium and acetic acid portions of potassium acetate upon heating to 130° or higher, the acetic acid being lost through volatilization. This would leave the potassium ion to produce a base reaction.

C. Temporary Mounts

Spores or structures with mucilaginous coverings should never be moistened with alcohol followed with potassium hydroxide and then heated. Measurements following this standard quick-mount procedure are highly variable. This is true even in different mounts of teliospores from the same sorus.

To determine the reason for the erratic differences, a study was conducted on parts of the procedure. A composite of spores from several sori was divided into three samples. One sample was refluxed for 15 minutes in 95% ethyl alcohol, another sample was refluxed for 15 minutes in 4% potassium hydroxide, and the last sample was refluxed for a like period in 4% alcoholic potash (4 grams of potassium hydroxide in 100 ml 95% ethyl alcohol).

Spores in mounts from the sample refluxed in alcohol alone looked normal (FIG. 1A). Mounts of the sample refluxed in potassium hydroxide produced spores whose reticulations were considerably swollen and whose sheaths were disorganized (FIG. 1B). Spores in mounts from the alcoholic potash sample were devoid of sheath (FIG. 1C and 1D).

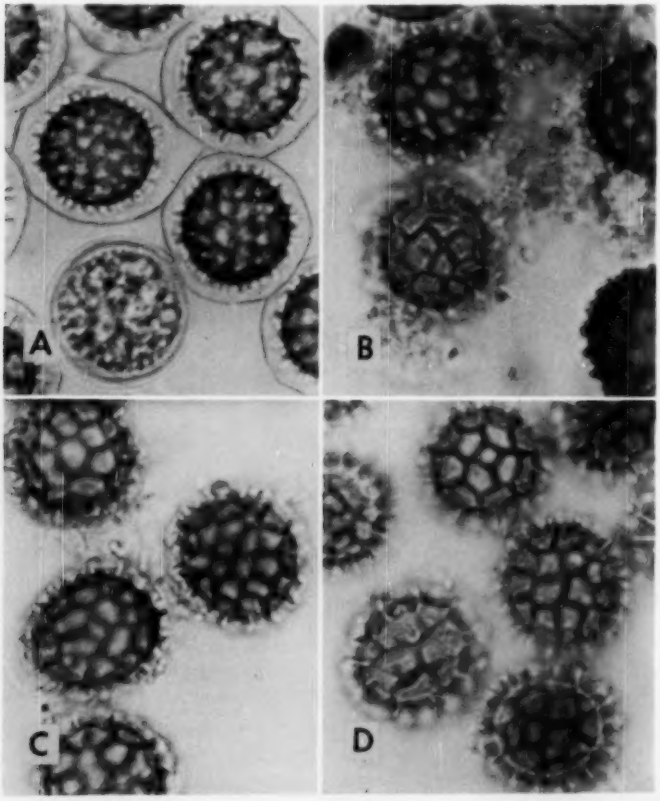


FIG. 1. The appearance of *T. contraversa* teliospores after refluxing for 15 minutes in: A. 95% ethyl alcohol with no apparent effect; B. 4% potassium hydroxide, causing disorganization of the sheath; C. 4% alcoholic potash which dissolved away sheath (median view of spores); and D. 4% alcoholic-potash (surface view of spores).

Thus, the standard quick-mount procedure invalidates observations on such morphologic structures as the sheath, because the sheath dissolves in hot alcoholic potash.

It is suggested that Shear's mounting fluid be adopted as standard for all quick mounts involving the smuts whether or not they possess a sheath or sheath-like structures. The medium is cheap, easy to prepare, produces clear mounts, and stores well in glass-stoppered bottles for several years without changing character.

One of the motives for employing the alcohol-potassium-hydroxide schedule in quick mounts is to leach away melanin pigments for finer definition of structures. It has been observed that a far better procedure is to oxidize these pigments to colorless compounds and then to follow such treatment with Shear's mounting fluid. However, such a schedule is satisfactory only where morphologic structures *per se* are to be considered. Strong oxidizing agents disorganize the protoplast and distort, unless the material has been adequately fixed in Karpechenko's solution (**7**) or some other fixing solution.

The procedure suggested when precise taxonomic measurements are not involved is to mount materials on a slide in 1% sodium hypochlorite (20% of commercial Clorox solution) for one minute, add Shear's mounting fluid and heat to drive off the alcohol, chlorine and water. A very clear, depigmented mount is obtained.

D. Suggested Standard Procedures in Taxonomic Studies on Smut Teliospores

As a result of the above observations it is suggested that the following procedures be employed in preparing teliospore mounts for taxonomic studies on the smuts.

1. Use a buffered Shear's mounting fluid of the following formulation:

2% potassium acetate in pH 8 McIlvaine's buffer (0.2 M)	300 ml
Glycerine	120 ml
Ethyl alcohol (95%)	180 ml

2. Mount spores on a glass slide in two drops of the buffered medium, fix with a coverslip and heat the slide over an alcohol lamp to remove volatiles. The mount is ready to evaluate.

For preparing semipermanent mounts, Chupp's double-coverslip method is recommended (**2**). The method essentially places all the spores on a common plane, and hence facilitates photography by minimizing depth-of-focus troubles. Being sealed, there is less trouble and worry from cleaning and storing reference mounts. Furthermore, slight

jars of the coverslip on unsealed mounts which cause spores to migrate are avoided.

TEIOSPORE-WALL RESPONSES TO DYES

The uptake of dyes by dwarf-bunt teliospores is erratic in aqueous mounts. Perhaps this is to be expected, since fungus walls in general have this characteristic. Since aqueous mounts are essential to morphological studies on smut teliospores, aqueous stains are also essential. Additional complications arise in manipulating aqueous stains for counterstaining. Many of them are incompatible. Following accepted schedules, most of the common dyes employed to differentiate specific structures will stain other parts. This results in poor definition of certain structures of smut teliospores. Another contributing variable is that some dyes cause sheath and reticular materials to contract. Only by following exacting procedures with selective combinations of dyes can these complications be avoided.

A. Pretreatment for Uniform Dye Uptake

In seeking a means to predispose spores for more uniform dye fixation, it was noted that leaching them for an hour in fat solvents such as ether, xylene, carbon tetarachloride, etc., would almost completely overcome the inconsistency of dye uptake. Such treated spores were not altered physically in any visible manner, and they absorbed most aqueous stains with ease. The leachates all contained relatively large amounts of fats. Apparently free or loosely-bound lipoidal materials impregnate the spore-wall layers in the smuts (and undoubtedly most fungi) and interfere with stain fixation from water carriers.

It was noted that much lower dye concentrations could be used after leaching. The contributing factor of dye astringency was lessened accordingly.

Thus, ether-leached spores were used in all staining tests employed unless otherwise indicated.

B. Responses of Wall Parts to Certain Dyes

Generally, stains in concentrations that are absorbed adequately by sheath material usually mask other wall layers. The sheath material usually stains the same color as the reticulum. However, certain dyes stain the sheath one color and the reticulum or other parts another. One of these is crystal violet.

Crystal violet stains sheath material adequately (lavender) at a concentration of about 0.1% or stronger. However, these concentra-

tions overstain the reticulum and inner spore materials to an opaque blue. At these concentrations the dye is also somewhat astringent on expanded sheath material. At a concentration of 0.01%, the dye stains the reticulum a light blue and the protoplast a navy blue, but it does not stain nor contract expanded sheath. However, it was observed that if weak alkali was added to the 0.01% dye, it intensified the blue of the reticulum, caused sheath to stain lavender-blue, and often differentiated the thick inner spore wall as dark red. The protoplast was stained a dense navy blue. The stain at the 0.01% concentration prepared with 0.2 M pH 8 McIlvaine's buffer caused the same effects.

Since mounts were not clear, attempts were made to adapt the dye to a similarly buffered Shear's mounting fluid. These attempts failed, owing to complete loss of the stain when the mount was heated to drive off volatiles. If buffered Shear's mounting fluid was added to the edge of a previously stained mount, considerable color was lost. If glycerine was added in concentrations above 50%, spores often collapsed; if added in lower concentrations, considerable color was lost.

The only method that gave satisfactory results was to prepare spores in Shear's mounting fluid according to standard procedures (including heating), but without adding a coverslip, adding pH 8 buffered stain, and then allowing the mount to desiccate without heat before adding the coverslip. A 0.01% solution of crystal violet in pH 8 buffered Shear's mounting fluid may be substituted for the buffered stain alone.

Differentiation of wall parts with other selective dyes combined with low concentrations of crystal violet were attempted. The only noteworthy dye of those dyes screened was rose bengal. In general, rose bengal at a concentration of 0.1% stains sterile cells and the protoplasts of immature teliospores a bright pink. It does not stain mature teliospores adequately except at much stronger concentrations, which are astringent. Crystal violet at 0.01% does not stain protoplasm strongly. As a reagent useful to define teliospore maturity and to accentuate sterile cells, rose bengal was investigated in combination with crystal violet. The two dyes are totally incompatible in solution. Both are precipitated upon mixing. However, if spores were prepared in Shear's mounting fluid according to standard procedures, stained with 0.01% crystal violet in buffered Shear's mounting fluid and flushed with dye-free mounting fluid to remove unabsorbed dye, 0.1% rose bengal in buffered Shear's mounting fluid could then be added to the mount. The result was satisfactory for defining immature spores and sterile cells; both were stained a bright pink. In mature spores the sheath stained pink and the reticulum stained a lavender-blue. In addition, the endospore

wall remained ochre-brown and the protoplast stained a blue-purple. Upon desiccation without heat to remove volatiles, there was no measurable distortion. Some interaction between the two dyes obviously was brought about.

The vital stain brilliant cresyl blue is readily absorbed by the reticulum and the protoplast and has no apparent astringency on sheath material, even in relatively strong concentrations (0.1%). Thus, it may well be considered as a suitable vital stain in germination studies of smuts or other fungi. Most of the other stains tested that are said to be vital stains were somewhat astringent to sheath and reticulum.

At a critical concentration, brilliant cresyl blue produced excellent differentiation of wall components. At approximately 0.01% the sheath stained pink, the reticulum stained bright blue, the endospore remained ochre-brown, and the protoplast stained an opaque blue. Below this critical level the sheath remained colorless and above the critical level the sheath and reticulum both were stained a dark brilliant blue. Attempts to define the specific concentration effecting these phenomena were abandoned, since every sorus tested seemed to vary in response to a specific dye concentration. All attempts to buffer or precondition spores for standardizing dye uptake failed. The critical level may be associated with relative spore maturity.

The dye nile blue A, at a concentration of 0.1% in unbuffered Shear's mounting fluid, defined a red band between the reticulum and the heavy endospore on spores that had not been leached in ether. Ether-leached spores did not show this red band, indicating the presence of an oily "mesospore" layer between the reticulum and the endospore. The dye stained sheath material light blue and the reticulum a brilliant blue. However, it also caused artifacts. Most notable were the lobations it produced on many spores. Sheath material seemed to mound-out of the areolar recesses, producing a phenomenon similar in appearance to the undulated surface of a raspberry. In addition, the sheath of many spores often showed well defined bands, increasing in color intensity the more proximal to the base of the reticulum. There was no positive reaction of the dye for "neutral" lipoids (i.e., only phospholipids or free fatty acids present at most) in either the sheath or the reticulum. The dye sudan black B weakly defined this oily "mesospore," but sudan III would not. Oil red was inconsistent.

SUMMARY

Owing to their composition, the teliospore walls of the smut fungi require hydrous mounting media for microscopic observations. *Tilletia*

contraversa Kühn possesses a mucilaginous sheath. It is differentially sensitive to the dehydrating tendencies and to the chemical nature of diverse reagents used in formulating various mounting media. Some reagents are astringent, and others over-expand structures. The sheath tends to over-expand under alkaline conditions and to contract variably under acid conditions. The reticulum, on the other hand, is not greatly affected by pH. Both the sheath and the reticulum are dilated by oxidizing agents.

Of the several mounting media tested, Shear's mounting fluid was the best. This medium gave clearer mounts and produced the least distortion, although there was some variability owing to pH changes during mount preparation. This variable was controlled by preparing the medium with pH 8 McIlvaine's buffer. The method of mount preparation and the formula of the medium are presented as a suggested standard procedure for taxonomic studies on smuts.

The standard temporary-mount procedure involving alcohol followed by potassium hydroxide and then heat should be discontinued, as hot alcoholic potash dissolves away sheath materials. Instead, Shear's mounting fluid should be employed.

To obtain uniform dye uptake by smut spores in aqueous mounts they should be leached in ether or some other highly volatile lipoid solvent. Not only are dyes more readily absorbed, but they are absorbed at much lower concentrations. Several staining procedures and dye combinations are suggested to differentiate wall structures. However, an oil-bearing "mesospore" could be defined only in non-ether-leached spores.

WASHINGTON STATE UNIVERSITY
PULLMAN, WASHINGTON

LITERATURE CITED

1. **Chupp, C.** 1940. Further notes on double cover-glass mounts. Mycologia **32**: 269–270.
2. **Chupp, C.** 1941. Diehl's double cover-glass mounts. Chron. Bot. **6**: 226–227.
3. **Diehl, W. W.** 1940. Mounting fluids and double cover-glass mounts. Mycologia **32**: 570–571.
4. **Fischer, G. W.** 1953. Manual of the North American smut fungi. The Ronald Press. New York, N. Y. 343 pp.
5. **Fischer, G. W., and E. Hirschhorn.** 1945. The Ustilaginales or "smuts" of Washington. State Coll. Wash. Agric. Exp. Sta. Tech. Bull. No. 459.
6. **Johansen, D. A.** 1940. Plant microtechnique. McGraw-Hill Book Co., Inc. New York. 523 pp.
7. **Rawlins, T. E.** 1933. Phytopathological and botanical research methods. John Wiley and Sons, Inc. New York. 156 pp.

FILAMENTOUS FUNGI FROM BLOSSOMS, OVARIES, AND FRUIT OF PICKLING CUCUMBERS [1, 2]

F. L. Raymond,[3] J. L. Etchells,[4] T. A. Bell,[5] and P. M. Masley [6]

(with 7 figures)

For the last two decades, cooperative investigations into fermentations of various foods in brine and silage have been carried on by the U. S. Department of Agriculture and North Carolina State College (**19**). Those studies have been especially concerned with fermentation problems of commercial packers of cucumber pickles.

The main problem, which remained unresolved until the advent of the present studies on the filamentous fungi, was that of producing firm pickles. In spite of carefully executed sanitation and brining procedures, many packers were consistently unable to produce pickles of highest-quality firmness. Furthermore, in certain years, a number of them experienced the loss, through softening of stock, of the contents of some or all of the vats filled during the early part of the brining season.

The literature bearing on the bacteriology of the brine-fermentation of cucumbers in relation to enzymatic softening has recently been thoroughly reviewed by Demain and Phaff (**9**). They concluded that the much-studied aerobic bacilli, with their high-pH-optima pectinolytic enzyme systems, appear to play only a minor role in softening. In this

[1] Cooperative investigations of the Southern Utilization Research and Development Division, Agricultural Research Service, United States Department of Agriculture, with the Department of Horticulture of the North Carolina Agricultural Experiment Station, and with the senior author and the late W. Lawrence White of Harvard University, who died shortly after these studies were begun.

[2] Published with the approval of the Director of Research, North Carolina Agricultural Experiment Station, as Paper No. 903 of the Journal Series.

[3] Formerly Research Assistant, Harvard University; now Research Officer, Forest Biology Division, Research Branch, Canada Department of Agriculture, to which organization he is grateful for time made available to complete this manuscript.

[4] Head, U. S. Food Fermentation Laboratory, ARS, USDA.

[5] Associate Chemist, U. S. Food Fermentation Laboratory, ARS, USDA.

[6] Formerly Graduate Assistant, North Carolina State College; now Senior Resident in Medicine, St. Vincent's Hospital, New York, New York.

connection, the acid and salt content of curing brines—as well as their low oxygen tension—would provide highly unfavorable conditions for the development of aerobic bacilli; essentially the same would be true for the soft-rot bacteria.

The low pH optima (4.0) for pectinolytic enzymes associated with softening of commercially brined cucumbers (**4, 6**) would point to fungi as the enzyme source rather than to bacteria. Although yeasts are highly acid- and salt-tolerant and are active in cucumber fermentations (**2, 10, 11, 12, 16, 18**), enzymological studies on a large collection of isolates from brines gave no evidence that they would be incriminated as a potential source of the brine-stock softening enzyme, pectinase (**3**). Therefore, it was logical to assume that the softening enzymes known to be active in brines (**4, 6**) were introduced with the cucumbers rather than produced in the brine during fermentation.

Enzymatic studies of various parts of the cucumber plant (*Cucumis sativus* L.) disclosed the presence of pectinolytic enzymes in staminate and pistillate flowers (**1, 5**) although the specific causative agent(s) responsible for the enzyme activity was not determined. It was further observed (**13, 14**) that about five times as many early-harvested cucumbers retained their blossoms as did the fruit harvested later in the season. Since blossoms already had been indicated as possible carriers of the active softening enzymes, the specific source appeared to lie with microorganisms borne by them. A series of studies were initiated at the beginning of the 1952 season to investigate populations, identity and enzyme production of filamentous fungi associated with pickling cucumbers and to further substantiate the hypothesis that the requisite enzymes were chiefly of fungal origin.

Demonstrations (a) that blossoms do carry the softening agents into commercial vats and (b) that filamentous fungi can produce those agents have been presented earlier (**13, 14**). Brinings of cucumbers from which the blossoms had been detached produced firm pickles; when those blossoms were included, the resulting pickles were soft. Adding to the normal complement of retained blossoms further increased softening. A simple, inexpensive, but effective softening-enzyme control measure was evolved which consists of draining off the original cover brine 36–48 hours after the vats are filled and replacing it with a new brine (**13, 14**).

The present paper deals primarily with the isolation, classification, and frequency of filamentous fungi on blossoms, ovaries, and fruit of cucumbers from commercial fields, a greenhouse, and a commercial brining station. The remaining aspects will be subjects of other papers.

MATERIALS AND METHODS

Samples of cucumber blossoms, ovaries, and fruit (FIG. 2) were collected at weekly intervals from a commercial brining station at Ayden in eastern North Carolina, from five field locations within 20 miles of the station, and from a greenhouse at North Carolina State College, Raleigh (FIG. 1). Samples from the brining station consisted of cucumber fruit typical of stock being sent in by receiving and grading

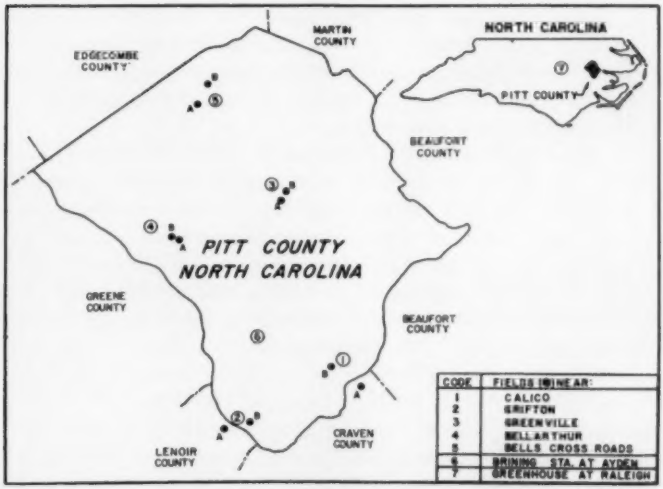


FIG. 1. Map showing origin of the samples; sampling locations are explained in the legend. Field samples were collected from the "A" fields except that the "B" fields were the source for samples collected in the last three periods at field location 1 and in the final period at field location 5.

stations which supply the cooperating company, and of blossoms which had been retained by the fruit. Greenhouse samples were of newly opened pistillate perianths and of the ovaries which supported them. Field samples, in addition to the fresh blossoms and ovaries, also included fruit typical of that being harvested for brining. Information about the collections is summarized in TABLE I.

Aseptic precautions were observed throughout. All samples were placed in sterilized paper bags by means of sterile forceps. At the end of each day of collecting, the samples were refrigerated until the fol-

lowing morning, at which time they were processed to produce inocula for culturing.

Each blossom sample, after being weighed, was blended in 0.85% saline for one minute at 14,000 rpm in a sterile, 50 ml Omnimixer[7, 8] blending-chamber. For the fresh blossoms, the dilution employed was 1:10 (w/v); however, a 1:50 dilution was found more convenient for the desiccated perianths collected at the station.

TABLE I

ORIGIN, NATURE, AND WEIGHT OF SAMPLES COLLECTED

Source	Type	Units per sample	Average weight (g)		Weekly samplings		Total
			Sample	Unit	Starting date	Ending date	
Five commercial fields[a]	Blossoms[c]	25	3.51	0.140	6/5	7/17	7
	Ovaries	25	12.00	0.480	6/5	7/17	7
	Fruit[d]	20	265.4	13.3	6/12	7/10	5
Brining station[a]	Blossoms[c]	45–100	0.5–2.0	0.017	6/12	7/10	5
	Fruit[d]	20	313.8	15.7	6/12	7/10	5
Greenhouse[b]	Blossoms[c]	25	4.15	0.166	6/26	7/17	4
	Ovaries	25	9.68	0.387	6/26	7/17	4

[a] Packer variety of cucumbers.
[b] Model variety of cucumbers.
[c] Perianths of freshly-opened, pistillate flowers.
[d] No. 1 size, 1 to 1¼ inches in diameter.
[e] Detached from the fruit at time of sampling.

Weighed samples of ovaries were shaken in 125 ml Erlenmeyer flasks with equal weights of saline for 10 minutes on a rotary shaker[8, 9] at 210 oscillations per minute.

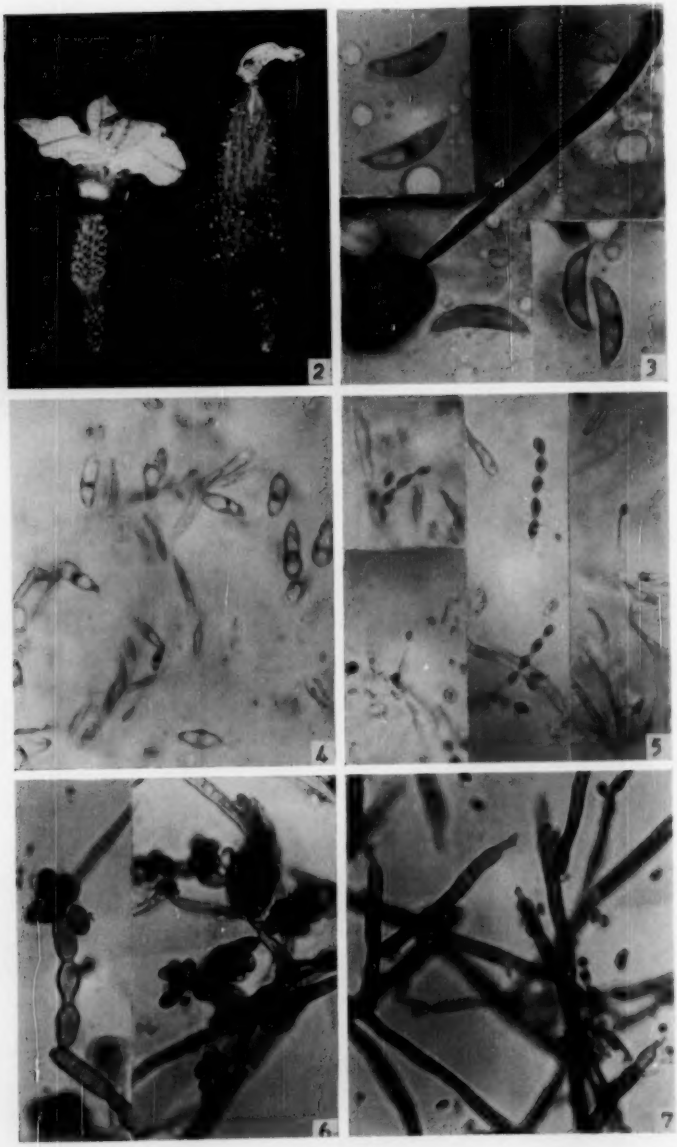
Fruit samples were similarly diluted but were placed in one-liter flasks which were shaken 100 times by hand.

Inoculation was accomplished by withdrawing and streaking on agar plates 0.01-gram aliquots of several different dilutions of each inoculum by means of a calibrated platinum loop. The media used for the isolation of filamentous fungi were Littman's oxgall agar with streptomycin (**20**) and Wickerham's nitrogen-base broth (**30**) with glucose, acidified

[7] Manufactured by Ivan Sorvall, Inc., Norwalk, Connecticut.

[8] It is not the policy of any of the organizations represented by the authors to recommend the products of any company over similar products of any other company. The name is supplied for informational purposes.

[9] Model V, capacity 40 flasks, manufactured by the New Brunswick Scientific Co., New Brunswick, New Jersey.



FIGS. 2–7.

to pH 3.5–3.7 with tartaric acid and solidified with agar, as previously described (**17**). Both media facilitate isolation of fungi by restricting the spread of most colonies and inhibiting the growth of many bacteria.

After incubation, usually for four days at 28° C, the sets of plates which had received the most extreme dilutions containing fungi were selected, the colonies were counted and recorded, and isolations were made by transferring bits of mycelia directly from the colonies to potato-dextrose-agar slants. This medium was used to maintain the cultures and to grow them for identification purposes. Other media employed in identification were Czapek's agar for the Aspergilli and Penicillia (**22**, **27**) and de Vries's gallic-acid agar in the case of *Cladosporium cladosporioides* (Fres.) de Vries (**28**).

Each of the dilutions used for inoculation was tested for pectinolytic activity. Also, many of the subsequently isolated fungi were investigated for pectinolytic activity, and some for cellulolytic activity. The methods employed were those used by Bell et al. (**3**, **4**, **6**) and by White and Downing (**29**), respectively. The results are discussed in detail in a companion paper devoted to populations and softening enzyme activity of fungi from the cucumber plant (**15**). However, summarized information for the action of the various identified species on pectin and for certain species on cellulose, based on the above report, is included herein.

RESULTS

NATURE AND OCCURRENCE OF FILAMENTOUS FUNGI

A total of 1032 isolates of filamentous fungi was obtained. These represented at least the 83 different entities listed in TABLE II. The number will vary according to the systems of classification employed; e.g., if the isolates representing the genus *Fusarium* had been segregated in accordance with the species-concept of Wollenweber and Reinking

FIGS. 2–7. FIG. 2. Left: the blossom and ovary of a freshly-opened pistillate flower of the cucumber (*Cucumis sativus* L.); right: a No. 1 size (1 to 1¼-inch diameter) fruit with retained perianth. The illustrated structures are typical of those collected in the samplings. Approx. 0.5 ×. FIGS. 3–7. Unidentified sporulating fungi; magnification 800 ×. FIG. 3. Conidia and seta of unidentified sporulating fungus A. FIG. 4. Unidentified sporulating fungus B. The distinctive but variable spores and the spindle-shaped "hyphal" cells can be seen. FIG. 5. Chains of conidia and two single phialides of unidentified sporulating fungus C. FIG. 6. Conidiophores of unidentified sporulating fungus D with spore-heads borne by phialides in the second (left) and third (right) types of configuration mentioned in the text. FIG. 7. Several conidiophores of unidentified sporulating fungus E. Note the peculiar, hyaline spore-bearing tips.

TABLE II

NAMES, NUMBERS, FREQUENCY, SEASONAL DISTRIBUTION, AND PECTINOLYTIC ACTIVITY OF THE FUNGI ISOLATED

Organisms	No. of isol.	Frequency of isolation from all samples					Seasonal distribution	Pectinolytic activity
		Sampling by weeks			by media[a]			
		Fields (35)[b]	Station (5)[b]	Greenhouse (4)[b]	LO (41)[b]	NB (41)[b]		
PHYCOMYCETES								
Choanephora cucurbitarum (B. & R.) Thaxt.	4	3	1	—	3	1	ML[c]	—
Cunninghamella echinulata (Thaxt.) Thaxt.	1	—	1	—	1	—	M	N[d]
Mucor sylvaticus Hagem	19	5	4	—	9	4	EM	P
Mucor sp. 2	1	1	—	—	—	1	E	P
Mucor sp. 3	1	1	—	—	1	—	E	P
Rhizopus arrhizus Fischer	1	1	—	—	—	1	E	P
R. stolonifer (Ehrenb. ex Fr.) Lind	4	—	2	—	1	1	EL	P
ASCOMYCETES								
Chaetomium globosum Kunze	2	—	—	2	2	—	M	N
Pseudonectria sp.	1	1	—	—	—	1	E	P
Rosellinia vel aff.	1	1	—	—	—	1	E	N
Thielavia basicola (B. & Br.) Zopf	3	3	—	—	2	1	E	P
FUNGI IMPERFECTI[e]								
Alternaria teuuis auct.	88	31	2	4	29	23	EML	P
Ascochyta cucumis Fautr. & Roum.	159	35	4	—	34	37	EML	P

[a] The media are Littman's oxgall agar (LO) and Wickerham's nitrogen-base agar (NB).

[b] The figures in parentheses indicate the total number of times which an entity could possibly occur in that category.

[c] The symbols E, M, and L refer to Early, Middle and Late portions of the harvest according to the following sampling weeks: E = the first two; M = the third through the fifth; and L = the last two.

[d] Representative cultures were screened for ability to bring about glycosidic hydrolysis of pectin; N means negative; P, positive; NP indicates that some of the tested strains gave negative results, others positive; — means not tested.

[e] Most of these organisms undoubtedly represent Ascomycetes but were placed here because no perfect-stage fruiting was found. None of the sterile fungi had clamp connections.

TABLE II—*Continued*

Organisms	No. of isol.	Frequency of isolation from all samples					Seasonal distribution	Pectinolytic activity
		Sampling by weeks			by media[a]			
		Fields (35)[b]	Station (5)[b]	Greenhouse (4)[b]	LO (41)[b]	NB (41)[b]		
Ascochyta sp.	1	1	—	—	1	—	L	N
Aspergillus elegans Gasp.	1	—	—	1	1	—	L	P
A. fumigatus Fres.	7	7	—	—	1	6	EML	P
A. niger Tiegh.	1	1	—	—	1	—	M	P
A. sydowi (Bain. & Sart.) Thom & Ch.	1	—	—	1	1	—	L	P
A. terreus Thom	1	—	1	—	—	1	M	P
A. unguis (Em.-W. & G.) em. Thom & Raper	1	1	—	—	1	—	E	P
A. versicolor (Vuill.) Tirab.	2	1	—	—	1	1	E	P
Cephalosporium sp. 1	5	3	1	—	4	—	EML	NP
Cephalosporium sp. 2	1	1	—	—	1	—	E	P
Cercospora sp.	1	1	—	—	—	1	M	N
Cladosporium cladosporioides (Fres.) de Vr.	104	31	5	3	37	13	EML	P
Colletotrichum lagenarium (Pass.) Ell. & H.	8	7	1	—	8	—	EML	P
Curvularia trifolii (Kauff.) Boed.	4	3	—	1	3	1	ML	P
Fusarium episphaeria (Tode) em. S. & H.	1	—	—	1	—	1	L	P
F. moniliforme Sheld. em. S. & H.	10	6	2	—	5	4	EML	P
F. oxysporum Schl. em. S. & H.	60	19	5	0	19	18	EML	P
F. roseum Lk. emend. S. & H.	106	27	5	1	31	12	EML	NP
F. solani App. & Wr. em. S. & H.	58	25	4	—	21	18	EML	P
Fusarium sp.	14	10	2	2	5	9	EML	—
Geotrichum sp.	3	—	2	—	2	—	M	N
Gliocladium roseum Bain.	2	2	—	—	2	—	M	N
Helminthosporium sp.	2	2	—	—	—	2	ML	N
Heterosporium terrestre Atk.	1	1	—	—	1	—	E	P
Humicola fusco-atra Traaen	1	1	—	—	1	—	E	N
Metarrhizium anisopliae (Metsch.) Sor.	2	2	—	—	—	2	EM	P
Myrothecium roridum Tode ex Fr.	4	3	—	—	3	—	L	NP

TABLE II—*Continued*

Organisms	No. of isol.	Frequency of isolation from all samples					Seasonal distribution	Pectinolytic activity
		Sampling by weeks			by media[a]			
		Fields (35)[b]	Station (5)[b]	Greenhouse (4)[b]	LO (41)[b]	NB (41)[b]		
M. verrucaria (Alb. & Schw.) Ditm. ex Fr.	9	8	—	—	8	—	EML	N
Myrothecium sp.	2	2	—	—	2	—	ML	NP
Papularia arundinis (Cda.) Fr.	1	1	—	—	1	—	E	N
Penicillium atramentosum Thom	1	1	—	—	—	1	M	P
P. chermesinum Biourge	1	1	—	—	—	1	E	P
P. duclauxi Delacr.	2	1	1	—	—	2	L	P
P. expansum Lk.	8	0	3	—	1	3	EML	P
P. frequentans Westl.	1	—	—	1	—	1	L	P
P. funiculosum Thom	2	—	—	2	—	2	ML	P
P. gladioli Mach.	1	—	1	—	—	1	M	P
P. herquei Bain. & Sart.	1	1	—	—	1	—	E	P
P. implicatum Biourge	1	—	1	—	—	1	L	P
P. janthinellum Biourge	16	10	1	—	4	9	EM	P
P. nigricans (Bain.) Thom	5	3	—	—	—	3	E	—
P. oxalicum Curr. & Thom	162	29	5	1	30	31	EML	P
P. piscarium Westl.	2	2	—	—	2	—	EM	P
P. restrictum Gilm. & Abb.	2	2	—	—	—	2	EM	P
P. rugulosum Thom	1	—	1	—	—	1	M	P
P. steckii Zal.	11	1	4	1	2	5	EM	P
P. sublateritium Biourge	7	5	—	—	4	3	EML	P
P. thomi Zal.	2	1	—	—	1	—	L	P
P. urticae Bain.	1	—	—	1	1	—	M	P
P. variabile Wehm.	2	1	1	—	1	1	M	P
Penicillium sp.	1	1	—	—	1	—	E	P
Pestalotiopsis macrotricha (Kleb.) Stey.	3	2	—	—	—	2	EL	P
Phoma sp.	7	2	5	—	1	6	ML	P
Pullularia pullulans (de Bary) Berkh.	3	1	1	—	—	2	E	P

TABLE II—*Continued*

Organisms	No. of isol.	Frequency of isolation from all samples					Seasonal distribution	Pectinolytic activity
		Sampling by weeks			by media[a]			
		Fields (35)[b]	Station (5)[b]	Greenhouse (4)[b]	LO (41)[b]	NB (41)[b]		
Sporotrichum pruinosum Gilm. & Abb.	3	2	—	1	1	2	EM	P
Stagonospora sp.	1	—	—	1	—	1	M	P
Stysanus sp.	1	—	—	1	1	—	L	N
Trichoderma viride auct.	16	11	1	—	6	7	EML	NP
Truncatella sp.	1	1	—	—	—	1	E	P
Undet. sporulating fungus A	28	17	—	—	17	—	EML	—
Undet. sporulating fungus B	15	5	1	—	—	6	E	—
Undet. sporulating fungus C	2	2	—	—	—	2	M	N
Undet. sporulating fungus D	2	2	—	—	—	2	EL	P
Undet. sporulating fungus E	1	1	—	—	—	1	M	N
Undet. sterile fungus A	11	10	—	—	8	2	EML	NP
Undet. sterile fungus B[f]	5	4	—	—	—	4	ML	N
Undet. sterile fungus C	2	2	—	—	—	2	EM	N
Undet. sterile fungus D	1	1	—	—	1	—	E	P
Undet. sterile fungus E	1	1	—	—	1	—	M	P

[f] The information given for this entity is incomplete; it was believed not to be a filamentous fungus and cultures were isolated only from plates of the sampling date 6/26/52.

(**31**) instead of the Snyder and Hansen concept (**24**, **25**, **26**), the list would have been much longer. Identification of the *Fusaria* was facilitated by the fact that the collection assembled by the late Dr. W. Lawrence White of Harvard University contained many cultures which had been determined by Sherbakoff or Snyder. Also, the authors are indebted to Dr. C. W. Hesseltine of the Northern Utilization Research and Development Division (USDA), Peoria, Illinois, whose provision of numerous strains of *Penicillium* greatly assisted in the determinations for this genus.

The following is a brief discussion of isolates by genera in alphabetical order:

Alternaria. All of these isolates were taken to represent the single species *A. tenuis* auct., *sensu lato*. This was one of the two most ubiquitously and abundantly distributed fungi found. However, it was encountered in only two of the five sampling weeks at the brining station. Strong pectinolytic activity and high frequency both indicate that this entity is important as a source of softening enzymes in samples of cucumber material collected at the brining station (cf. **15**).

Ascochyta. All but one of the 160 isolates were assignable to *A. cucumis* Fautr. & Roum., which is the imperfect stage of *Mycosphaerella citrullina* (Smith) Gross., the widespread attacker of cucumber vines and fruit. Despite the fact that this entity was absent from greenhouse samples, it occurred with greater frequency than any other fungus in the study. Isolates tested were strongly or very strongly pectinolytic. This organism is considered to be another major source of softening enzymes observed for cucumber material (cf. **15**).

Aspergillus. Members of this genus, consisting of 14 isolates representing seven species, were considered of too limited occurrence to be very important in softening. The pectinolytic activity of most species was moderate.

Cephalosporium. These isolates proved weakly pectinolytic and were notable only for the fact that none was derived from the nitrogen-base medium.

Cercospora. The single isolate showed no pectinolytic activity.

Chaetomium. The two isolates of *C. globosum* Kunze were from greenhouse samples; they were strongly cellulolytic but negative for action on pectin.

Choanephora. The four isolates of the cucumber blossom-blight organism, *C. cucurbitarum* (Berk. & Rav.) Thaxt., died before they could be tested for action on pectin; however, this fungus occurred too rarely to be important here.

Cladosporium. All of these isolates were placed in *C. cladosporioides* (Fres.) de Vries, a species morphologically similar to *C. cucumerinum* Ell. & Arth. Twenty-five of them were examined by Prof. N. N. Winstead at North Carolina State College, and were excluded by him from *C. cucumerinum* on the basis of pathogenicity tests on cucumber and tomato plants. De Vries's gallic-acid test similarly excludes them.

This organism is the other of the two most ubiquitously and abundantly distributed fungi isolated. Like *Alternaria tenuis,* it occurred in all five of the field samples; unlike that organism, it was isolated from the brining-station samples for all of the five sampling weeks. It was obtained almost three times as frequently on Littman's agar as on the nitrogen-base medium.

Cultures tested for pectinolysis were strongly or very strongly active. This fungus was undoubtedly one of the more important sources of softening enzymes found for cucumber material (cf. **15**).

Colletotrichum. Only one of the eight isolates of *C. lagenarium* (Pass.) Ell. & Halst., the anthracnose organism, was obtained from brining-station samples. For this reason, and also because of its comparative infrequency, it is considered to be of minor importance as a source of pectinolytic enzymes, despite strong activity.

Cunninghamella. The single isolate of *C. echinulata* (Thaxt.) Thaxt. was negative for pectinolysis.

Curvularia. C. trifolii (Kauff.) Boed. occurred too rarely to be important here even though it was strongly pectinolytic.

Fusarium. Most of the 249 isolates represented one or another of the three most common species, *F. oxysporum* Schl. emend. Snyder & Hans., *F. roseum* Lk. emend. Snyder & Hans., and *F. solani* App. & Wr. emend. Snyder & Hans. Those three species were of frequent occurrence except from greenhouse samples.

Thirty isolates, representing the five species listed in TABLE II, were tested for pectinolytic activity; all but one was positive. Members of this genus obviously are important as a potential source of softening enzyme activity observed for samples of material (cf. **15**).

Geotrichum. All three isolates were pectinolytically negative.

Gliocladium. G. roseum Bain.; the single isolate tested was negative for pectinolytic activity.

Helminthosporium. The two isolates tested were strongly positive for pectinolysis but this entity was too rare to be important here.

Humicola. The single isolate of *H. fusco-atra* Traaen showed moderate pectinolytic activity.

Metarrhizium. Both isolates of *M. anisopliae* (Metsch.) Sor., the "green muscardine" which attacks many insects, were pectinolytically negative. This fungus was present in the samples from one field for two consecutive weeks. Although its presence is readily explained by assumption that infected insects were associated with the plants sampled, and although it clearly is unimportant here, it should be pointed out that published information shows that this organism can be an active saprophyte.

Mucor. All but two of these isolates were typical of *M. sylvaticus* Hagem. Five isolates in this species were tested for pectinolysis and all were found moderately active. Because of this and the fact that its frequency at the station was relatively high, this species may be a moderately important contributor of softening enzymes.

Myrothecium. Two of the 11 isolates tested were weakly pectinolytic; the remainder were negative. Although members of this genus apparently contribute very little as sources of pectinolytic softening enzymes, they may be important in any cellulolytic softening.

Of the 15 isolates representing this genus, all but two were readily assignable to *M. verrucaria* (Alb. & Schw.) Ditm. or *M. roridum* Tode ex Fr. The other two had spores which were similar to those of *M. roridum* but much longer.

Papularia. The single isolate of *P. arundinis* (Cda.) Fr. was considered negative for pectinolytic activity.

Penicillium. Although all of the isolates which were tested proved strongly positive for pectinolytic activity, only those representing *P. oxalicum* Curr. & Thom were considered sufficiently abundant to be of substantial importance as a source of softening enzymes. Seven isolates of this species were tested for cellulolytic activity and all were able to reduce the tensile strength of cotton tape by 90 or 95%. A culture of *Myrothecium verrucaria* and one of *Chaetomium globosum,* which were selected for comparison, caused 100% and 90% loss of strength, respectively.

As can be seen from TABLE III, *P. oxalicum* was one of the more abundant fungi in the field samples; of even greater significance is the fact that it was the most abundant in the brining-station samples. There is little reason to doubt that this species was an important contributor to the concentration of pectinolytic enzymes found in samples of cucumber material, particularly flowers, during the 1952 season (cf. **15**).

The frequency of occurrence of *P. janthinellum* Biourge, *P. steckii* Zal. and *P. expansum* Lk. in field and station samples was high enough so that they are considered of moderate importance.

Pestalotiopsis. *P. macrotricha* (Kleb.) Stey. occurred too rarely to be of importance and was only weakly pectinolytic.

Phoma. The seven isolates assigned here probably represent a single species. They do not represent *Macrophomina phaseoli* (Maubl.) Ashby, the charcoal-stem-rot fungus. The three cultures tested were positive for pectin degradation.

TABLE III

FREQUENCY (%) OF OCCURRENCE OF THE SEVEN MOST IMPORTANT FUNGI IN SAMPLES FROM THREE SOURCES

Organism	Frequency of isolation from samples collected at:						
	Brining station		Commercial fields			Greenhouse	
	Blossoms %	Fruit %	Blossoms %	Ovaries %	Fruit %	Blossoms %	Ovaries %
Penicillium oxalicum	90	70	86	71	100	—	17
Cladosporium cladosporioides	50	20	71	64	90	38	17
Fusarium roseum	50	60	64	43	70	12	—
Fusarium oxysporum	60	70	57	36	80	—	—
Fusarium solani	30	70	50	71	80	—	—
Ascochyta cucumis	60	40	93	100	100	—	—
Alternaria tenuis	10	30	93	86	70	25	33

Pseudonectria. The single isolate is noteworthy for the fact that the imperfect stage belongs to *Fusarium;* no such relationship has been reported previously for these genera. This fungus has been described recently as *P. diparietospora* by Miller et al. (**21**).

Pullularia. *P. pullulans* (de Bary) Berkh. occurred infrequently; the three isolates tested were positive for action on pectin.

Rhizopus. Although all of the isolates of *R. stolonifer* (Ehrenb. ex Fr.) Lind and *R. arrhizus* Fischer were active pectinolytically, they were of such limited occurrence that they must be considered to have only minor roles as contributors to enzyme softening of cucumbers.

Rosellinia, vel aff. The single isolate was pectinolytically negative.

Sporotrichum. Isolates of *S. pruinosum* Gilm. & Abb. were rarely obtained and only weakly pectinolytic.

Stagonospora. The single isolate was pectinolytically positive.

Stysanus. The one culture was negative for pectinolysis.

Thielavia. *T. basicola* (B. & Br.) Zopf, represented by three isolates, was pectinolytically weak.

Trichoderma. *T. viride* auct. was common enough from field samples to warrant consideration. However, only one isolate in every three was positive for action on pectin and none was present in station blossom

samples, and so it was concluded that this entity is of only minor importance here.

It was interesting to note that during the first four sampling weeks, eight of the ten cultures were isolated from Littman's agar, while all six obtained during the final three weeks were derived from the nitrogen-base medium. No corresponding shift in morphological characteristics was found.

None of the isolates manifested the loose, tuftless mycelial growth or the *Gliocladium*-like conidiophores which frequently occur in strains that produce gliotoxin or viridin, and which Miller et al. (**21**) recently described as *Gliocladium virens*.

Truncatella. The single isolate was pectinolytically positive.

Unidentified sporulating fungi. Of the next five entities discussed, several generic names are known for one of them but not accepted, whereas the genera to which the other four belong are "new" to these writers. Because an entity unknown to some often is known to others, the authors prefer not to diagnose five new genera. The five are here illustrated and briefly described with the hope that readers who may be familiar with them or are working with similar organisms will be benefitted by having this information. Cultures will be available upon request.

Unidentified sporulating fungus A. This interesting fungus probably is relatively unimportant as a source of softening enzymes. Although it was moderately abundant in the field samples, it was never obtained from station or greenhouse samples. All isolates came from Littman's agar.

The cultural appearance of this fungus is similar to that of *Verticillium albo-atrum;* i.e., there is a paucity of aerial hyphae, and the dominant feature is the black aspect presented by myriads of small, black sclerotial bodies on the surface of the agar. Microscopically, however, there is less similarity; short, black setae occur among the sclerotia, and the numerous curved, hyaline spores, suggestive of *Menispora,* frequently become several-septate at maturity (FIG. 3).

Unidentified sporulating fungus B. This organism has been the subject of discussion among the authors of this paper. Some regard it as a filamentous fungus, others would classify it among the yeasts. The spores are quite unyeastlike but the mycelium can be interpreted as being composed of spindle-shaped cells which form false hyphae with false branching. FIGURE 4 illustrates the variable spores and some hyphal cells which have been slightly disoriented by pressure on the coverslip.

The 15 isolates of this fungus came from cucumber material representing only one of the sampling periods. It was present, however, during most or all of them and was abundant during these investigations. All of the isolates were obtained from the nitrogen-base medium.

Unidentified sporulating fungus C. This fungus is congeneric with the imperfect state of *Cephalotheca sulfurea* Fuckel [10] (8) and with certain of the fungi which were described as species of the untenable genus *Spicaria* and which more recently have been transferred to *Paecilomyces* (7). This latter disposition is not accepted because the fungi included in that concept of *Paecilomyces* can be segregated into at least three groups which are distinguished by their phialides and conidia. A paper in which a more satisfactory disposition will be proposed is now in preparation.

Characteristic of the group which the fungus represents are the tapering narrow-tipped phialides and, particularly, the chains of spores connected by extremely narrow isthmi (FIG. 5).

Unidentified sporulating fungus D. This entity exhibits a pattern of sporulation which has superficial similarities to that of *Cladosporium* but which results in heads rather than chains of spores. The greatest similarity is in the elliptical phialide cells which may arise singly on undifferentiated aerial hyphae and sporulate immediately, which may proliferate into chains of such cells and terminate in spores, or which may be organized on verticillately branched conidiophores (FIG. 6). Both conidia and phialides remain unicellular with smooth, light-brown walls.

Unidentified sporulating fungus E. This fungus also has a distinctive method of sporulation. The conidiophores are stout, erect, simple or branched, and with dark-brown walls. They are terminated by abruptly narrowed, irregularly flexed hyaline structures which bear the conidia in the manner of a *Tritirachium* in which each stalk is so short that its spores are compacted into a ball. Similar structures usually develop subsequently from near the apices of one or more of the subterminal cells of each conidiophore (FIG. 7).

Undetermined sterile fungi. None of these had the clamp connections which would have indicated that they were Basidiomycetes. Of the five entities in this section, only sterile fungus A was of more than rare occurrence and it was isolated only from field samples. Three of four cultures of this entity proved pectinolytically positive. It is considered to be of minor importance here.

[10] Chesters's culture was obtained from the Centraalbureau voor Schimmelcultures, Baarn, Netherlands.

COMPARISON OF ISOLATION MEDIA

Of the 83 entities listed in TABLE II, 28 were obtained only from Littman's agar, and 29 only from the acidified nitrogen-base agar. Of the 26 fungi which occurred on both media, seven were more common on the one, eight were more common from the other, and eleven occurred in approximately equal proportions on both agars. It must be pointed out, however, that most of the entities occurred too infrequently to justify the drawing of conclusions from only the figures given above. It is necessary to consider those organisms which were abundant.

Of the ten most common fungi which were obtained from both field and station samples, two occurred more frequently on one medium, two on the other, and six were about equally frequent on both.

It must be concluded, therefore, that the two isolation media were about equally effective in permitting enumeration of the common members of the fungal flora of pickling cucumbers. If either had been used alone, the same entities would have been found abundant—although possibly in a slightly different order of frequency—but the list of rare fungi would have been smaller. For the purpose of designating the important fungal sources of softening enzymes, either medium would have been satisfactory. However, if isolation were to be repeated for that purpose, the nitrogen-base agar would be selected; on it, the fungal colonies remained small and restricted much longer (several days) than was the case with Littman's agar.

DISCUSSION AND CONCLUSIONS

The 83 entities of fungi to which the isolates were assigned are easily divided into two categories based on their importance as potential contributors to enzymatic, particularly pectinolytic, softening of cucumber material. The occurrence of an organism in station samples is probably of greater significance than is its abundance in field samples, because of the fact that the softening enzymes, resulting chiefly from fungal growth, are carried into the vats predominantly on blossoms retained by the cucumbers. The fungi which are considered to be of major importance as sources of softening enzymes in cucumber material are those which were common in both station and field samples; those which occurred infrequently obviously belong in the category of minor or no importance.

TABLE III lists, in what is presumed to be their approximate order of importance, the seven species which are considered to have been major contributors to softening enzyme activity of cucumber brines in

eastern North Carolina during the 1952 season. The great frequency of *Penicillium oxalicum* in brining-station blossom samples necessitates placing this potent fungus high in the list. *Cladosporium cladosporioides* and the three common species of *Fusarium* are also placed high, for the reason that those four fungi were found to be about 1000 times as abundant on station blossoms as on field blossoms. The order of increase in propagative units on station perianths over field perianths was 200 times for *Ascochyta cucumis* and 500 times for *Alternaria tenuis*. It is possible that much or most of the increase in propagules resulted from sporulation as the blossoms dried on their way to the station. Although differences between sporulating and non- or lightly-sporulating fungi should have been minimized by the blending, they still may have been great enough to have caused these last two entities to be placed lower in the list than they belong. Also, it is realized that there may be differences in enzyme concentrations between spores and hyphal cells, or, for that matter, in the amount of enzyme secreted into the blossom by fungal growth. The list is arranged on the basis of frequency and *in vitro* enzyme potency and is subject to rearrangement when additional information becomes available.

The other category comprises the rest of the species isolated; these are of minor importance as individual sources of brine-stock softening enzymes but they still may have a role, if only a small one, as contributors to softening activity. More than half of the minor species were positive when tested for pectinolytic activity and most are known (**15**, **23**) to produce cellulolytic enzymes.

It is believed that the origin of softening enzyme activity of commercial cucumber brines under the conditions described is brought about by enzymes contributed by filamentous fungi which enter the vats chiefly on blossoms retained by the cucumbers. Many fungi contribute to softening but the seven species listed in TABLE III stand out as being the most important.

SUMMARY

Results are presented for the identification of fungi isolated from samples of blossoms, ovaries and immature fruit of the cucumber (*Cucumis sativus* L.) collected throughout one season from five commercial fields, a brining station and a greenhouse, all of which were located in North Carolina. Of the 1032 cultures isolated, 964 (93.4%) were reduced to 73 species in 34 genera; the remaining 68 isolates (6.6%), representing 10 distinct entities, were not fully identified. Thirty-one species in five major genera represented 80.50% of the total isolations;

42 species in 29 minor genera accounted for 12.9%. Seven species in the five major genera—*Penicillium, Ascochyta, Fusarium, Cladosporium* and *Alternaria*—represented 71.4% of all isolations. The species were: *Penicillium oxalicum*, 15.7%, of isolates; *Ascochyta cucumis*, 15.4%; *Fusarium roseum*, 10.3%; *Cladosporium cladosporioides*, 10.1%; *Alternaria tenuis*, 8.5%; *Fusarium oxysporum*, 5.8%; and *Fusarium solani*, 5.6%. These species comprised most of the total fungus populations of field blossoms and station blossoms collected throughout the season and were considered to be an important source of pectinolytic and cellulolytic enzymes.

FOREST BIOLOGY LABORATORY
SOUTHERN RESEARCH STATION
MAPLE, ONTARIO
AND
U. S. FOOD FERMENTATION LABORATORY
AND NORTH CAROLINA STATE COLLEGE
RALEIGH, NORTH CAROLINA

LITERATURE CITED

1. **Bell, T. A.** 1951. Pectolytic enzyme activity in various parts of the cucumber plant and fruit. Bot. Gaz. **113**: 216–221.
2. ——, **and J. L. Etchells.** 1952. Sugar and acid tolerance of spoilage yeasts from sweet-cucumber pickles. Food Technol. **6**: 468–472.
3. ——, **and** ——. 1956. Pectin hydrolysis by certain salt-tolerant yeasts. Appl. Microbiol. **4**: 196–201.
4. ——, —— **and I. D. Jones.** 1950. Softening of commercial cucumber salt-stock in relation to polygalacturonase activity. Food Technol. **4**: 157–163.
5. ——, —— **and** ——. 1951. Pectinesterase in the cucumber. Arch. Biochem. Biophys. **31**: 431–441.
6. ——, —— **and** ——. 1955. A method for testing cucumber salt-stock brine for softening activity. U. S. Dept. Agr. *ARS-72-5*. 15 pp.
7. **Brown, A. H. S., and G. Smith.** 1957. The genus *Paecilomyces* Bainier and its perfect stage *Byssochlamys* Westling. Brit. Mycol. Soc. Trans. **40**: 17–89.
8. **Chesters, C. G. C.** 1935. Studies on British Pyrenomycetes. I. The life histories of three species of *Cephalotheca* Fuck. Brit. Mycol. Soc. Trans. **19**: 261–279.
9. **Demain, A. L., and H. J. Phaff.** 1957. Cucumber curing: softening of cucumbers during curing. Jour. Agr. Food Chem. **5**: 60–64.
10. **Etchells, J. L.** 1941. Incidence of yeasts in cucumber fermentations. Food Research **6**: 95–104.
11. ——, **and T. A. Bell.** 1950. Classification of yeasts from commercially brined cucumbers. Farlowia **4**: 87–112.
12. ——, —— **and I. D. Jones.** 1953. Morphology and pigmentation of certain yeasts from brines and the cucumber plant. Farlowia **4**: 265–304.

13. ——, —— **and** ——. 1955. Studies on the origin of pectinolytic and cellulolytic enzymes in commercial cucumber fermentations. Food Technol. **9**(3): 14, 16. 1955 (Abstr.).
14. ——, —— **and** ——. 1955. Cucumber blossoms in salt stock mean soft pickles. N. C. Agr. Expt. Sta., Research & Farming **13**: 14–15.
15. ——, ——, **R. J. Monroe, P. M. Masley, and A. L. Demain.** 1958. Populations and softening enzyme activity of filamentous fungi on flowers, ovaries, and fruit of pickling cucumbers. Appl. Microbiol. **6**: 427–440.
16. ——, **R. N. Costilow and T. A. Bell.** 1952. Identification of yeasts from commercial cucumber fermentations in northern brining areas. Farlowia **4**: 249–264.
17. ——, ——, —— **and A. L. Demain.** 1954. Control of molds during the enumeration and isolation of yeasts from soil and plant material. Appl. Microbiol. **2**: 296–300.
18. ——, **and I. D. Jones.** 1943. Bacteriological changes in cucumber fermentation. Food Indus. **15**(*2*): 54–56.
19. **Goresline, H. E.** 1942. Cooperative pickling investigations of the U. S. Department of Agriculture. Fruit Prod. Jour. & Amer. Vinegar Indus. **21**: 232–233.
20. **Littman, M. L.** 1947. A culture medium for the primary isolation of fungi. Science **106**: 109–111.
21. **Miller, J. H., J. E. Giddens and A. A. Foster.** 1957. A survey of the fungi of forest and cultivated soils of Georgia. Mycologia **49**: 779–808.
22. **Raper, K. B., and C. Thom.** 1949. A manual of the *Penicillia*. Williams & Wilkins, Baltimore. 875 pp.
23. **Reese, E. T., H. S. Levinson, M. H. Downing and W. L. White.** 1950. Quartermaster culture collection. Farlowia **4**: 45–86.
24. **Snyder, W. C., and H. N. Hansen.** 1940. The species concept in *Fusarium*. Amer. Jour. Bot. **27**: 64–67.
25. ——, **and** ——. 1941. The species concept in *Fusarium* with reference to section *Martiella*. Amer. Jour. Bot. **28**: 738–742.
26. ——, **and** ——. 1945. The species concept in *Fusarium* with reference to *Discolor* and other sections. Amer. Jour. Bot. **32**: 657–666.
27. **Thom, C., and K. B. Raper.** 1945. A manual of the *Aspergilli*. Williams and Wilkins, Baltimore. 373 pp.
28. **de Vries, G. A.** 1952. Contribution to the knowledge of the genus *Cladosporium* Link ex Fr. Uitgeverij & Drukkerij, Baarn. 121 pp.
29. **White, W. L., and M. H. Dowing.** 1951. *Coccospora agricola* Goddard, its specific status, relationships, and cellulolytic activity. Mycologia **43**: 645–657.
30. **Wickerham, L. J.** 1951. Taxonomy of yeasts. U. S. Dept. of Agr. Tech. Bull. *1029*. 56 pp.
31. **Wollenweber, H. W., and O. A. Reinking.** 1935. Die Fusarien. Parey, Berlin. 355 pp.

SOME VIEWPOINTS ON THE PHYLOGENY OF RUST FUNGI. III. ORIGIN OF GRASS RUSTS [1]

E. E. Leppik

(WITH 3 FIGURES)

Serious economic losses caused by cereal rusts to our grain crops and field grasses are well known facts of much concern to plant growers. For example, it is estimated that six species of the genus *Puccinia*, which attack our main cereals, are causing more losses every year than all other rusts combined (Arthur, **1929**, p. 326). This is due to the fundamental value of these crops, which form the main basis of the food supply for both man and domestic animals, and also to the wide-spread and aggressive nature of the rusts involved. Therefore, it is no wonder that the cereal rusts have been the object of most intensive study for centuries.

In spite of the importance and the increasing scientific interest of the grass rusts, little is known about the phylogeny and evolution of this group of fungi. In most existent monographs and manuals, referred to at the end of this paper, little information is given about the origin and genetic relationships of grass rusts. Up to the present time, no generally accepted phylogenetic system has been proposed either for taxonomic classification or for practical identification of the enormous number of rusts on gramineous plants.

Yet our most effective control measure, the breeding of rust-resistant varieties, depends ultimately upon our knowledge of the genetic constitution and phylogenetic relationships of the rusts involved. Through a series of genetic studies on the flax rust, Flor (**1955**, **1956**) has recently shown that the host and parasite possess complementary genetic systems, which are based upon a series of multiple alleles at several gene loci. Any gene for resistance in the host plant acts only when there is a cor-

[1] Reports I and II of this series were published in Mycologia **45**: 46–74. 1953, and **48**: 637–654. 1956.

Journal Paper No. J-3526 of the Iowa Agricultural and Home Economics Experiment Station, Ames, Iowa. Project No. 1073. Disease Reports of the NC-7 Regional Plant Introduction Station, U. S. Department of Agriculture, Ames, Iowa. No. 2.

responding gene in the parasite for virulence. It is convincingly evident that the present-day epidemic rusts have evolved from their relatively less aggressive ancestors on certain wild grasses by means of a selective adaptation to our over-cultivated grain crops and food grasses. Every man-bred wheat or corn variety developed for high food value offers also a new chance for a rapidly expanding rust race, unless the complementary genetic system for rust resistance has been improved correspondingly. It is from this point of view that the phylogenetic study of grass rusts appears almost imperative to the plant pathologist in charge of disease-resistance work.

PHYLOGENY OF GRASS RUSTS

A predominant number of rust fungi on gramineous plants belong to the closely related large genera *Puccinia* and *Uromyces*. These two genera, in their turn, include nearly one-half of all the known species of the Uredinales. Four additional genera on tropical grasses, *Physopella* (*Angiopsora*), *Dasturella*, *Phacopsora*, and *Stereostratum*, bear only little weight, insofar as their species number and geographic distribution are concerned. In view of their morphological characteristics, which are melampsoroid (*Physopella*), ochropsoroid (*Dasturella*), cronartioid (*Phacopsora*), and gymnosporangioid (*Stereostratum*), these tropical rusts might well be regarded as special phylogenetic groups, evolved from some ancestral conifer rusts before the main expansion of the Puccinieae (Fig. 3). The present-day grass rusts, consequently, appear to be of polyphyletic origin at their tribal level, but they all may be readily derived from ancestral conifer rusts of the large family Melampsoraceae sensu Dietel (**1928**).

Most earlier authors, attempting to surmount the difficulty of the insufficient phylogenetic knowledge of the fungi, have arranged all known species of the genera *Puccinia* and *Uromyces* according to their host plants. This means of classification, in spite of its obvious practical advantage for species identification, is almost inapplicable for phylogenetic purposes. The great number of hosts which most rusts in this group inhabit, and the many grass species that bear more than one rust species at a time make the whole system undesirably artificial and unfeasible phylogenetically.

Arthur (**1906**) developed his own system for the classification of rust fungi, dividing *Puccinia*, *Uromyces* and other genera into numerous smaller units, mainly according to their complete or incomplete life cycles. This method appeared so radically different from all previous systems that all material had to be rearranged and genera redefined.

Arthur did this in the North American Flora (**1907–1927**). This purely artificial system, however, did not receive much recognition by other workers and was later abandoned by its founder. Contrary to his previous view, Arthur (**1934**) then united all his former puccinioid genera into a single genus *Puccinia*, but divided the new combination into two large sections: *Eupuccinia* and *Bullaria*. Although this method of grouping has been criticized by Dietel (**1928**, p. 91), Cummins (**1936**, p. 128) and others, no further systems have been proposed, except that Clements and Shear (**1931**) turned back to the old *Puccinia-Uromyces* order.

Thus little unanimity of opinion existed regarding the phylogeny and classification of grass rusts until the whole problem could be approached from an entirely different angle (Leppik, **1953**, **1956**; Savile, **1954**). The purpose of the present paper, however, is not the taxonomic classification of grass rusts, but rather an attempt at hologenetic grouping of these fungi according to their evolutionary sequence (see FIGS. 1, 2). For this purpose the morphological characteristics of species diagnoses are used as presented by Sydow (**1904–1915**), Arthur (**1919**, **1934**) and Cummins (**1953**, **1956a**, **1956b**).

SPECIES RELATIONSHIP AMONG GRASS RUSTS

In order to study the evolution of grass rusts, particularly their occasional jumping from one host group to another, more detailed information is needed about their species relationship and biological specialization. Since such information is not sufficiently available in the present taxonomic treatises, new methods must be employed.

For taxonomic purposes the genus *Puccinia* is commonly divided into a few subgenera or sections, which contain several hundred species in each subdivision. For instance, Arthur's (**1934**) two sections *Eupuccinia* and *Bullaria* contain 313 and 202 species respectively, on various host plants in the United States and Canada alone. It is evident that such a large number of species cannot be closely related genetically, but represent rather a certain evolutionary level of numerous parallel-evolving smaller groups. In the section *Eupuccinia*, Arthur (**1934**, p. 101) included various species characterized by oblong or clavate teliospores, firm persistent pedicels, and aecidioid aecia with verrucose aeciospores. Yet all these morphological characteristics point to a lower evolutionary level of the genus *Puccinia*, a primitive stage of development, much like the *Gymnotelium* stage of the genus *Gymnosporangium* (see below). The second section *Bullaria*, on the contrary, has globoid or ellipsoid teliospores with short, fragile pedicels and uredinoid aecia with echinu-

late aeciospores. These more advanced characteristics obviously have been reached by many parallel-evolving phylogenetic groups, which are specialized to live on higher angiosperms.

A further crucial point for the classification of grass rusts is the artificial genus *Uromyces*. Single-celled and bi-celled teliospores differ so clearly from one another that the genus *Uromyces* for one and *Puccinia* for the other appear to be most natural divisions among puccinioid rusts, and were long accepted as such by earlier authors. Yet the reduction in the number of cells of teliospores is a prevalent evolutionary trend among puccinioid rusts, which can be traced back to their gymnosporangioid ancestors. Many parallel-evolving phylogenetic groups of the genus *Puccinia* have reached this single-celled stage of their teliospores and are marked frequently by intermediate forms. Consequently the number of cells of the teliospores has no phylogenetic value at all and the genus *Uromyces* is disregarded by Arthur (**1934**) and other recent authors.

At the present stage of knowledge it is exceedingly difficult to arrange the huge number of species of the genus *Puccinia* into a compact phylogenetic system. As long as the species relationships and the historical development of the whole genus are not sufficiently known, the phylogenetic value of any tentative system would remain questionable. For hologenetic purposes, therefore, different methods of classification must be employed, and some new terms—sib, sept, clan, kin—introduced, as follows below.

In conventional taxonomic systems genera are divided into subgenera, subgenera into sections, sections into series, and so on, until closely related groups of species are established. However, instead of this descending system from genus down to species an opposite, ascending order from species up to genus is equally practicable. First, several closely related species can be united into small groups (septs), these into larger units (clans), and so on until some higher taxonomic order (section or series) is reached (see TAB. I).

The Anglo-Saxon word "sib" (sibb, gesib) is frequently used by geneticists for indication of close genetic relationship, as brothers and sisters, irrespective of sex. Plant breeders commonly indicate by this term descendents from interpollinated parents of the same pure line. In open-pollinated plants and in fungi sibs are practically impossible to recognize and the term is used mainly in artificial pollination and isolated cultures.

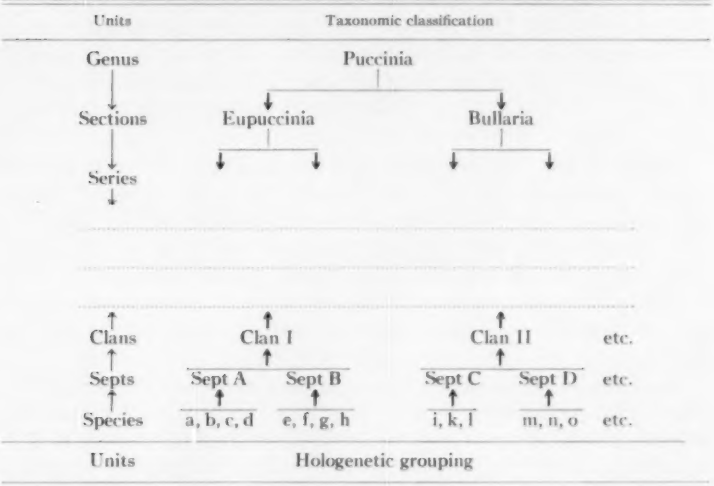
"Clan" is of Gaelic (clann) and "sept" of Irish origin. In some cases these words coincide with the Anglo-Saxon "kin" or "cynn" (see

Webster's International Dictionary). In their original languages all these words have almost the same meaning: offspring, descendents from a common ancestor, an exogamous division of a tribe, and so on.

In the present paper we want to indicate with the term "sept" natural groups of closely related species which descend from a common ancestral stock and show sufficient resemblance in their morphological characteristics and biological specialization.

TABLE I

PROVISIONAL CLASSIFICATION OF THE GENUS PUCCINIA INTO SECTIONS (ACCORDING TO ARTHUR, 1934) AND CLANS (SEE TEXT)



"Septs" are the smallest phylogenetic units, which evolve parallel to one another and extend sometimes through several hologenetic stages (see FIG. 1). For instance, the sept of the coronate rusts of the section *Eupuccinia* is easily distinguishable by the digitate projections on teliospores, and by its globoid, finely echinulate, polyporous urediospores. The core of this sept is formed by the closely related *Puccinia coronata* Cda., *P. coronifera* Kleb., and *P. himalayensis* (Barcl.) Diet. All these species live with their aecidial stage on *Rhamnus* and produce their telia and uredia on various grasses. From this ancestral "gene-center" about a dozen species of coronate rusts have migrated to various groups of grasses and higher angiosperms. Some other species, such as *P. mesnieriana* Thüm. and *P. schweinfurthii* (P. Henn.) Magn., were ob-

viously reduced to microcyclic forms and live now with their telia on *Rhamnus*. It should be noted here that several earlier authors, for example Arthur (**1906**), separated such microcyclic forms of *Puccinia coronata* into different genera. The correlation of these microcyclic species with *P. coronata* was first pointed out by Dietel in **1887** (inaug. diss. p. 25), whose viewpoint was later accepted by Arthur (**1934**, p. 154), Gäumann, and others.

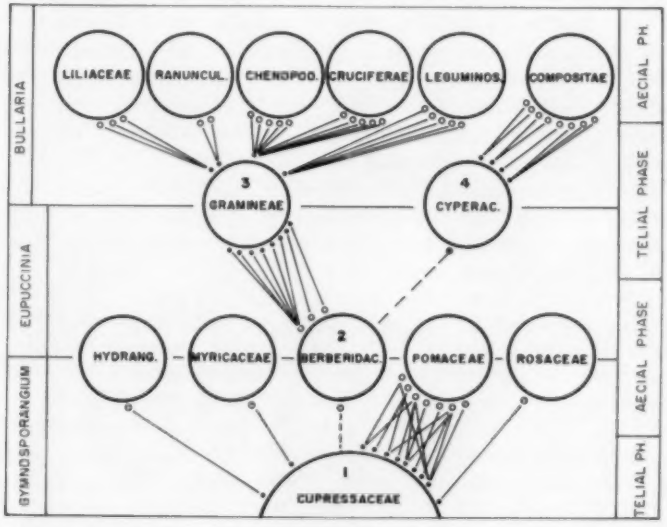


FIG. 1. The hologenetic sequence of biological specialization of *Gymnosporangium* and *Puccinia*. Black dots (●) indicate telia, small circles (○) aecia, which spore forms correspond to the sporophytic and gametophytic generations of these fungi. Notice the several "gene-centers" from which distribution of these fungi took place. For instance: 1. From Cupressaceae to many arboreous angiosperms. 2. From Berberidaceae to many grass groups and probably to Cyperaceae. 3. From Gramineae to many families of herbaceous angiosperms. 4. From Cyperaceae to Compositae. Assumable connections are indicated by dashes (———).

The sept of stem rusts of the same section, including *P. graminis* and related species, has spread from its "gene-center" on barberry to many grasses and higher angiosperms. Numerous members of this sept extend today through several hologenetic stages to the higher angiosperms. This sept also has its microcyclic forms in several stages.

When several related septs show sufficient resemblance in their main characteristics, they can be united into a "clan," the next ascending rank

above sept. In addition to morphological similarity, the members of the same clan are commonly specialized to live on some group of closely related host plants. Related clans can be further united into "kins" and so on, until taxonomic units such as series, or sections, are reached. In a previous report (Leppik, **1956**, p. 643–646), for instance, the 24 clans of the genus *Gymnosporangium* were provisionally called "paragraphs" and united into 10 sections and 2 subgenera.

But even in this provisional grouping of the genus *Puccinia* in TABLE I a considerable gap between clans and sections remains unfilled until more phylogenetic information is gathered about these fungi. Nevertheless, the introduction of these new terms and a new type of classification is unavoidable for the hologenetic grouping of grass rusts. However, "sept," "clan," and "kin" are not intended to be taxonomic units in the sense of the botanical code, but are used only with reference to the hologenetic approach. Analogous ecological terms for subspecific units are already in use, as follows: idiotype, ethnotype, ecotype, biotype, morphotype are ascending typological units which gradually lead to taxonomic classification: races, varieties, subspecies, species.

HETEROECISM AND AUTOECISM

It is still an unexplained and much discussed problem whether the autoecious or heteroecious forms are more primitive and ancestral among rust fungi. Cunningham (**1931**, p. 84) believed that heteroecism is an ancient and autoecism is a recent condition, which supposition opposed the earlier views held by Fischer (**1898**), Dietel (**1899**), McAlpine (**1906**), Christman (**1907**), and Olive (**1911**).

According to Fischer's theory (**1898**, p. 115), primitive rusts must necessarily have been autoecious and plurivorous because of their assumed saprophytic and unspecialized nature. He postulated, for instance, that *Puccinia coronata* originally had been able to live in both aecial and telial stages on grasses and on Rhamnales. Only later did this rust specialize itself for definite hosts and heteroecism. Fischer's theory, although generally accepted by earlier workers, has been criticized by Klebahn (**1904**, p. 171), Cunningham (**1931**, p. 86) and Leppik (**1948**, p. 3). Since the biological specialization appears to have been obligatory as far back as the entire group can be traced, *Puccinia coronata* must have been heteroecious from its very beginning.

Cunningham's view, on the contrary, rested mainly upon the fact that the ancient family Melampsoraceae is predominantly composed of heteroecious rusts, unlike the modern Pucciniaceae, which contains only

three heteroecious as against numerous autoecious genera. This evidence supports the belief that autoecism increases and heteroecism decreases, as the scale is ascended from the Melampsoraceae to the Pucciniaceae.

Most recent writers, Gäumann (**1928**, p. 591 ff., **1952**, p. 326), Dietel (**1928**, p. 36 ff.), and Savile (**1955**, p. 72, 78, 81 ff.), among others, consider the genera *Uredinopsis*, *Milesina*, and *Hyalopsora* as being the most primitive genera of all known rust fungi. But since all these and closely related genera of Pucciniastraceae are typically heteroecious, heteroecism seems to be, according to these authors, the primitive state and autoecism the more advanced condition among these fungi. Fischer's theory (l. c.) that the primitive rusts were autoecious and plurivorous obviously had little support from this quarter.

The present study, however, revealed several new facts and some fundamental concepts which seem to have a direct bearing on the further treatment of the problem. Application of the hologenetic methods enabled us to establish the relative phylogenetic age of various rust groups and their host plants in a way described elsewhere (Leppik, **1953**, **1956**). It appeared from this study that the genera *Uredinopsis*, *Milesina*, and *Hyalopsora* are not elementary but rather advanced types in the evolutionary sequence of rust fungi. Their telial hosts, the eusporangiate ferns of the northern hemisphere, are not the primary hosts, as suggested by the cited authors, but got their relatively advanced and well specialized rusts from conifer trees. Also, the typically heteroecious genus *Gymnosporangium*, contrary to the suggestion of Olive (**1911**, p. 145), Gäumann (**1928**, p. 592–593) and Dietel (**1941**, p. 356), has possibly arisen from the aecioid conifer rusts, shifting its aecial stage from Cupressaceae to Rosales, but not vice versa.

It is evident that all the above-mentioned writers, in searching for ancestral forms among present-day rusts, actually dealt with advanced types. Neither *Puccinia coronata* nor *Gymnosporangium* or *Uredinopsis* is primitive in regard to its complicated life cycle or highly specialized spore forms. These rusts represent the top achievements of several parallel-evolving phylogenetic trends on various host groups. The truly primitive stomatosporous and primarily autoecious rusts on tropical ferns of the type *Desmella* are erroneously joined in all present systems with the modern family of Pucciniaceae. The presumable origin and early evolution of the rust fungi, as much as can be concluded from the present study, are outlined briefly in a previous paper (Leppik, **1955**, p. 150–152).

Primitive rusts on ferns can be best derived from the ancient Auriculariales, which presumably had already lived in the Paleozoic era.

Several ancient forms among the Auriculariales, characterized by a marked similarity to primitive rusts, are still living in tropical ferns. *Herpobasidium filicinum* (Rostr.) Lind., *Platycarpa polypodii* Couch, and *Uredinella coccidiophaga* Couch are appropriately described as intermediate genera and species between the primitive rust fungi and Auriculariales. *Eocronartium muscicola* (Fr.) Fitzpatr. and *Iola javensis* Pat. exhibit some further similarities to primitive fern rusts.

These early rusts on ferns evidently differed from their ancestral Auriculariales mainly in their well established probasidia (elementary

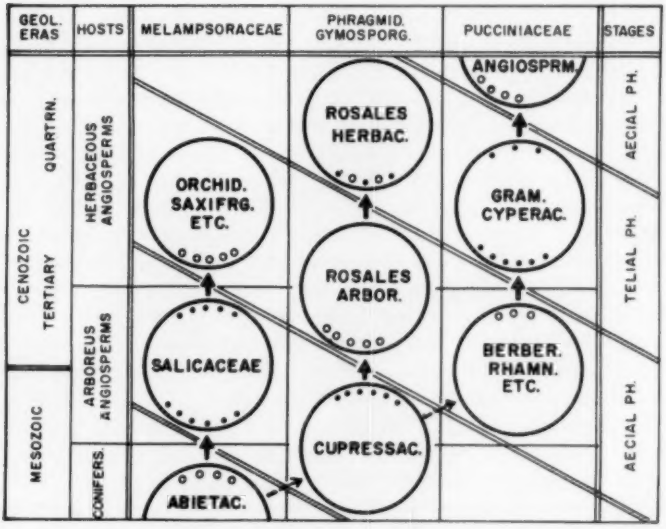


FIG. 2. The hologenetic sequence of three main rust groups, Melampsoraceae, Gymnosporangiae and Pucciniae, arranged according to their historical development and migration through different types of vegetation. Legends the same as in FIG. 1, explanation in text.

teliospores) and urediospores. Mycelium of these rusts has very probably been intercellular, similar to that of *Herpobasidium*, with well established haustoria and sporogenous fascicles emerging through stomata of affected fern fronds. Host-stimulating activity of these rusts must have been still low, not causing any hypertrophy in the host tissues. Several recent genera of primitive stomatosporous rusts, like *Desmella, Hemileia, Cystopsora, Gerwasia,* and others, exhibit similar characteristics and

could be united, therefore, with ancient fern rusts into a single group, Stomatosporales (Trib. Hemileieae Dietel, pro parte). From their striking similarity with primitive Auriculariales and absence of aecia this group could be considered as the most primitive among recent rust fungi.

There is no evidence that these primitive fern rusts had ever possessed an aecial stage or heteroecism. As a matter of fact, no aecium has ever been described on ferns and no heteroecism is known for the above-mentioned primitive stomatosporous rusts. Even higher fern rusts of the family Pucciniastraceae form their aecia regularly on conifers, but never on ferns.

In the light of these facts and findings, the futility of attempts to find the beginning of heteroecism in some secondarily adapted autoecious rusts of higher order may be now admitted. Both stages, heteroecism and autoecism, are fixed genetically with the alternation of generations, which is irreversible in its progression. Therefore heteroecious rusts cannot jump deliberately with both generations to new host groups, as erroneously postulated by Fischer (**1898**, p. 115). One generation must hold its old host group until the next generation is well established on a new group. Then the first generation disengages itself from the old hosts and shifts its corresponding phase to some phylogenetically younger group of plants, as new hosts (Leppik, **1953**, p. 70). Thus the heteroecious rusts with their two generations, telial stage (sporophyte), and aecial stage (gametophyte), climb the hologenetic ladder, step by step as sketched in FIG. 2.

BIOLOGICAL SPECIALIZATION OF GRASS RUSTS

Regardless of the controversial assumptions discussed above, all grass rusts are strictly specialized and predominantly heteroecious. Fischer's theory that *Puccinia coronata* might have been, in the beginning, autoecious and plurivorous is irreconciliable with verifiable facts (Leppik, **1948**). Cunningham's theory of the decreasing heteroecism is only partly valid for grass rusts.

The complete life cycle of grass rusts is characterized by three successive phases on two consecutive hosts, as described below. With their dikaryophase (production of urediospores) and diplophase (production of teliospores) these rusts are sharply restricted to grasses and sedges, but their haplophase (formation of aeciospores) occurs on a large number of flowering plants. Consequently, the aeciospores and urediospores

of these rusts can infect grasses or sedges only, whereas their basidiospores are able to invade numerous families of angiosperms, except grasses and sedges. A former belief that the tender germs of basidiospores cannot penetrate the thick and hard epidermis of grasses has not been supported by experimental study. The discovery of *Puccinia graminella*, which lives with both aecia and telia on grasses (see below), also indicates against such a simple mechanical explanation.

A proper arrangement of grass rusts according to their hologenetic sequence reveals to us some new conditions. As it appears from FIG. 1, grasses and sedges have retained during the expansion of Pucciniaceae an intermediate position between primitive and more advanced angiosperms. First they have absorbed some rust strains from primitive angiosperms, such as *Puccinia graminis* on *Berberis* and *P. coronata* on *Rhamnus*. These pioneer rusts have adapted themselves to live on numerous closely related grass species, multiplying enormously by splitting themselves into numerous microspecies and races. In spite of this variety of telial hosts, they have to complete their life cycle on *Berberis* or *Rhamnus*. Only a very few autoecious rusts in this group, such as *Puccinia graminella* (Speg.) Diet. et Holw., *Uromyces pencanus* Arth. et Holw., and probably *P. digna* Arth. et Holw., secondarily have withdrawn their aecia from the intermediate host and complete all phases now on *Stipa*.

In the second stage of their historical development, the newly-formed grass rusts began to invade various younger groups of angiosperms, in particular those flowering plants which lived in close proximity to grasses and sedges. They retained telia on grasses or sedges but shifted aecia to certain younger angiosperms. At that stage of development these rusts started to disengage themselves from their previous telial hosts, i.e. grasses and sedges. After this separation they became autoecious, or transferred secondarily their telia to some still younger group of flowering plants. As a result of this transference, many younger families of angiosperms, such as the Compositae, Scrophulariaceae, Leguminosae, and others are exceptionally rich in autoecious rusts. This condition is of course in agreement with Cunningham's view of decreasing heteroecism among Pucciniaceae. But there are also heteroecious rusts among younger angiosperms which form telia on dicotyledonous plants, instead of grasses or sedges (FIG. 1).

In this way the grass rusts could keep pace with the fast evolving groups of younger angiosperms and could expand rapidly over vast grassland areas throughout the world.

ORIGIN OF GRASS RUSTS

The lowest forms among grass rusts obviously are those which live with their aecial stage on the Berberidaceae, Rhamnaceae, or other arboreous families of primitive angiosperms. The view that barberry might be the primary host and grasses the secondary hosts of *Puccinia graminis* was accepted by Fischer (**1898**), Olive (**1911**, p. 140) and other earlier workers. Arthur (**1934**) recognized the primitiveness of these rusts, including them in the elementary section *Eupuccinia*, and placed them in his system before the more advanced *Bullaria* on higher angiosperms. Olive (**1911**, p. 145), however, considered all heteroecious rusts as starting their existence from some autoecious ancestral stock on the present gametophytic host. This supposition is, of course, not in accordance with the established facts, as mentioned above.

According to the present study, barberry and buckthorn occupy the lowest stages in the hologenetic host sequence of grass rusts, as pictured in FIG. 2. Next higher are grasses and sedges, which represent the telial stage, and still higher are the phylogenetically younger flowering plants for the aecial stage of these fungi. This sequence is natural and corresponds to the historical development of the biological specialization of grass rusts.

If we accept the fact that the aecial stage on Berberidaceae is the most primitive among recent grass rusts, the question arises about the origin of barberry rust. According to the hologenetic ladder the next lower step in this sequence should be a telial stage on some phylogenetically older group than primitive angiosperms. But there are no direct hologenetic connections between Berberidaceae and any plant group lower than barberry. A number of primitive aecioid and puccinioid rusts on Magnoliaceae, Anonaceae and Lauraceae do not show much resemblance to grass rusts but appear to be more closely related to the genus *Gymnosporangium*. Hologenetically these rusts represent the same evolutionary level as barberry rust.

On the other hand, a striking parallelism exists between the hologenetic sequence of grass rusts and that of the Melampsoraceae. The latter group has its younger aecial forms on higher angiosperms, its telial phase almost exclusively on Salicales (*Salix, Poplus*), and its lower aecial stage on Abietaceae (Leppik, **1953**).

Both families, Pucciniaceae and Melampsoraceae, belong to the same order of aecioid rusts, which very likely evolved from some common ancestral stock. As a matter of fact, the lowest known telial stage for the Melampsoraceae lives on leptosporangiate ferns; the ferns are, however, out of the hologenetic sequence of Pucciniaceae.

In the hologenetic sequence of aecioid rusts, there lies between ferns and barberry such an impassable broad gulf that the forcible attempt to derive the family Pucciniaceae directly from the fern rusts appears wholly unintelligible. There is no evidence that grass rusts have had any direct connection with ferns. But there are some indications in the hologenetic sequence of grass rusts which point to the Cupressaceae as the possible lowest telial stage for the Pucciniaceae. First, it should be noted that the phylogenetic relationship between the Pucciniaceae and Gymnosporangieae is almost unanimously accepted by recent authors, who join these tribes into the same family Pucciniaceae (Dietel, **1928**; Cunningham, **1931**; Arthur, **1934**; Gäumann, **1952**). This view is mainly supported by the striking morphological resemblance between the *Puccinia* and *Gymposporangium*. The structural similarity of these two genera is particularly impressive when the phylogenetically lower subgenera, such as *Eupuccinia* and *Gymnotelium*, are compared. Both these subgenera have teliospores with long gelatinous pedicels, cupulate aecia, subepidermal pycnia, and uredia without peridia. Such a precise resemblance of main sporulating organs cannot occur by mere accident, but points rather to the close phylogenetic relationship of both these groups.

Second, these genera occupy a successive position one after another in the hologenetic sequence of their biological specialization (see FIG. 2). There is considerable evidence, therefore, that the primitive members of the section *Eupuccinia* (the ancestors of *Puccinia graminis* might have been among them) were evolved from some older form of *Gymnotelium* or from the provisional *Proto-Gymnosporangium* on *Cupressaceae* (Leppik, **1956**, p. 638).

Ample evidence exists that the early ancestors of the present genus *Gymnosporangium* spread from their original coniferous hosts to several arboreous angiosperms, where their successors still live. For example, several recent species of *Gymnosporangium* still live with their teliospores on Cupressaceae but transfer their aecia to Pomaceae (order Rosales), Hydrangeaceae (order Rosales), Myricaceae (order Myricales), and Rosaceae (order Rosales). Additional autoecious genera, such as *Phragmidium* on Rosaceae, *Cumminsiella* on Berberidaceae, *Uropyxis* on Fabaceae, and other related groups, obviously descended from the same ancestral stock of *Proto-Gymnosporangium* (see FIG. 1).

This intensive distribution of the early Gymnosporangiae from their original source very likely took place in the Cretaceous period, which is characterized by a wide expansion of arboreous angiosperms in the northern hemisphere. At that time large areas were covered with rapidly expanding angiospermous vegetation in a climatic condition which

was not favorable for the outmoded conifers. The Cupressaceae were already in their decline and could not follow and compete with the youthful angiosperms. Rust fungi, on the contrary, found in angiosperms a fertile soil for their expansion but were in many cases disconnected from their old coniferous hosts. It became necessary for these rusts to reduce their life cycle to autoecism or to transfer their telia to some angiosperms. Some rust clans on Berberidaceae, for instance, transferred their telial stage on grasses, very likely because barberry and grasses lived closely together in the same grassland areas. A wide expansion of grassland vegetation during the Tertiary period doubtless must have favored the distribution of puccinioid rusts until they reached their present dominating position among recent Uredinales.

The presumable origin of the Gymnosporangieae from the common stock of aecioid conifer rusts is described in a previous paper (Leppik, **1956**, p. 646). Ancient conifer rusts very likely lived in the Mesozoic era in the northern conifer forests, which had fern undergrowth. The heteroecious life cycle of these rusts on ferns (teliospores) and conifers (aeciospores) could have easily existed in this environment. But the separation of Cupressaceae from the ancient conifers and their migration to more arid mountainous areas eliminated the less adaptable ferns from the new type of vegetation. Rusts on Cupressaceae, consequently, lost their telial hosts and had to reduce their life cycle to autoecism. Later on they could restore heteroecism with new hosts, as soon as primitive angiosperms began to appear in the neighborhood. Thus these ancient rusts began to move their aecia to various groups of primitive angiosperms, in the same way that fern rusts once resettled their aecia to the ancient conifers. The resettlement of fern rusts, however, caused the beginning of a new family Melampsoraceae, the dominant rust group of recent conifer forests. Similarly, the move of conifer rusts from the Cupressaceae to primitive angiosperms created a new family of Pucciniaceae, the dominant rust group of grassland areas.

SOME CONCLUSIONS

In this study an attempt is made to derive the grass rusts, *Puccinia* and *Uromyces,* from the ancient Gymnosporangiaceae on the early Cupressaceae. This assumption about the origin of grass rusts is substantially supported by their close phylogenetic relationship, as established between the recent genera of the *Gymnosporangium* and *Puccinia.* Further evidence in favor of this theory can be deduced from the striking resemblance between the main spore forms of the phylogenetically lower subgenera *Gymnotelium* and *Eupuccinia.* In the hologenetic sequence

of their biological specialization, the genera *Gymnosporangium* and *Puccinia* occupy successive positions, one after another, as pictured in FIG. 1.

In the light of the above mentioned facts and findings, evidence is strong that the splitting of ancient conifer rusts successively into the Melampsoraceae, Gymnosporangiaceae and Pucciniaceae took place in an early geologic time, presumably in the Mesozoic era. But it is also obvious that the later evolution of these main rust groups has been rather a parallel development on various host plants and in different environments, as pictured in FIG. 2.

The combination of phylogenetic and hologenetic methods of study enables us to approach the problem of the origin and evolution of grass rusts from two different angles: from the phyletic and holetic view-

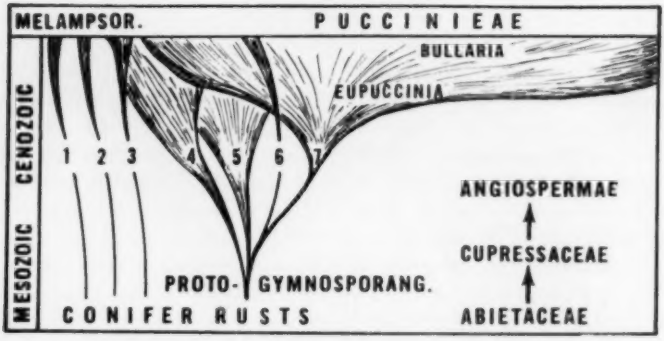


FIG. 3. Assumable phylogenetic roots of grass rusts. 1. *Physopella*. 2. *Dasturella*. 3. *Phacopsora*. 4. *Phragmidium*. 5. *Gymnosporangium*. 6. *Stereostratum*. 7. *Puccinia* and *Uromyces*.

points, as described elsewhere (Leppik, **1953**, p. 6). This is a highly satisfying stereoscopic picture, projected through phylogenetic and hologenetic lenses at the same time. One of its special beauties is that it shows us new fields, which were inaccessible with previous research methods, and clears up many former puzzles. On the basis of this new information, a natural system for phylogenetic classification of grass rusts might be worked out, as pictured in FIG. 3.

SUMMARY

According to the present theory, the early ancestors of the tribe Pucciniae have descended from the ancestral Gymnosporangiae, whose rusts in their turn have evolved from the ancient Melampsoraceae. Be-

ginning from an ancient stock of aecioid conifer rusts, these parasites moved first from the Abietaceae to Cupressaceae and changed their quarters from the ancient conifer forests to the medieval cupressus-angiosperm vegetation. In the new environment these fungi resettled themselves once more from the Cupressaceae to the arboreous angiosperms, inhabited thereafter grasses and sedges, and finally reached higher angiosperms. Their expansion began presumably from the early Mesozoic conifer forests of the northern hemisphere, continued through the mixed forests of the Cretaceous period, and terminated finally in the vast grassland areas of world-wide distribution.

ACKNOWLEDGMENTS

The author is greatly indebted to Dr. W. H. Bragonier, Head of the Department of Botany and Plant Pathology, Iowa State University, for his interest and help in this study. Dr. G. W. Martin of the State University of Iowa read the manuscript and offered some valuable suggestions.

Grateful acknowledgment is also made to the Manuscript Committee: Dr. J. C. Gilman, Chairman, Dr. R. W. Pohl, and Dr. M. D. Simons for their collaboration and critical remarks. Mrs. Lilian Feinberg assisted me in the preparation of the manuscript.

DEPARTMENT OF BOTANY AND PLANT PATHOLOGY
PLANT INTRODUCTION STATION
IOWA STATE UNIVERSITY
AMES, IOWA

LITERATURE CITED

Arthur, J. C. 1906. Eine auf die Struktur und Entwicklungsgeschichte begründete Klassification der Uredineen. Res. Sci. Congr. Bot. Vienne, 1905, 331–348.

——. 1919. Uredinales, in N. Am. Flora **7**(3): 18. 188.

——. 1929. Plant rusts (Uredinales). 1–446. John Wiley, N. Y.

——. 1934. Manual of the rusts in the United States and Canada. 438 pp. Lafayette, Indiana.

Bessey, E. A. 1952. Morphology and taxonomy of fungi. i–xiii, 1–791. Blakiston Co.

Clements, F. E., and C. L. Shear. 1931. The genera of fungi. 496 pp., 58 pl. H. W. Wilson & Co., New York; repr., Hafner Publishing Co., New York, 1957.

Christman, A. H. 1907. The alternation of generations and the morphology of the spore forms in the rusts. Bot. Gaz. **44**: 81–101.

Cummins, G. B. 1936. Phylogenetic significance of the pores in urediospores. Mycologia **28**: 103–132.

——. 1953. The species of *Puccinia* parasitic on the Andropogoneae. Uredineana (Encyclopédie Mycol. **24**) **4**: 1–90. P. Lechevalier, Paris.

——. 1956a. Uredinales parasitizing grasses of the tribe Chlorideae. Mycologia **48**: 126–162.

——. 1956b. Host index and morphological characterization of the grass rusts of the world. The Plant Disease Reporter, Suppl. 237, Washington, D. C.

Cunningham, G. H. 1931. The rust fungi of New Zealand. 261 pp. Dunedin, N. Z.

Dietel, P. 1887. Beiträge zur Morphologie und Biologie der Uredineen. Bot. Centrbl. **32**: 54–56, 84–91, 118–121, 152–156, 182–186, 217–220, 246–250.

——. 1903. Waren die Rostpilze in früheren Zeiten plurivor? Bot. Centralbl. **79**: 81.

——. 1928. Reihe Uredinales; in Engler und Prantl, Die Natürliche Pflanzenfamilien **6**: 24–98.

——. 1941. Bemerkungen über die geographische Verbreitung einiger Gattungen der Uredineen. Ann. Mycol. **39**: 350–359.

Fischer, Ed. 1898. Entwicklungsgeschichliche Untersuchungen über Rostpilze. Beitr. Kryptog. Schweiz **1**(10): 1–121.

Flor, H. H. 1955. Host-parasite interaction in flax rust—its genetics and other implications. Phytopathology **45**: 680–685.

——. 1956. The complementary genetic systems in flax rust. Advances in Genetics **8**: 29–59. Academic Press, New York.

Gäumann, E. 1926. Vergleichende Morphologie der Pilze. 1–626.

——. 1949. Die Pilze. Grundzüge ihrer Entwicklungsgeschichte und Morphologie. 382 pp. Birkenhäuser, Basel.

——. 1952. The fungi. 1–420. Hafner, N. Y.

——, **and C. W. Dodge.** 1928. Comparative morphology of Fungi. 701 pp. McGraw-Hill, New York.

Grove, M. A. 1913. The British rust fungi (Uredinales), their biology and classification. 412 pp. Cambridge University Press.

Klebahn, H. 1904. Die wirtswechselnden Rostpilze.

Leppik, E. E. 1948. Über die Pleophagie bei den Rostpilzen. Sendbrief Inst. Weihenstephan. 1–12.

——. 1953. Some viewpoints on the phylogeny of rust fungi. I. Coniferous rusts. Mycologia **45**: 46–74.

——. 1955. Evolution of angiosperms as mirrored in the phylogeny of rust fungi. Arch. Soc. Zool. Bot. Fenn. Vanamo **9** (suppl.): 149–160. Helsinki.

——. 1956. Some viewpoints on the phylogeny of rust fungi. II. Gymnosporangium. Mycologia **48**: 637–654.

McAlpine, D. 1906. The rusts of Australia, their structure, nature, and classification. 349 pp. Melbourne.

Olive, E. W. 1911. Origin of heteroecism in the rusts. Phytopathology **1**: 139–149.

Savile, D. B. O. 1955. A phylogeny of the Basidiomycetes. Can. Jour. Bot. **33**: 60–104.

——. 1954. Taxonomy, phylogeny, host relationship, and phytogeography of the microcyclic rusts of Saxifragaceae. Can. Jour. Bot. **32**: 400–425.

Sydow, Paul, and Hans Sydow. 1904, 1910, 1915. Monographia Uredinearum. Vol. I, II, III. Lipsiae.

THE AGARIC FLORA OF TEXAS. III. NEW TAXA OF BROWN- AND BLACK-SPORED AGARICS

HARRY D. THIERS [1, 2]

In previous papers (**5, 6**) descriptions were presented on new white- and pink-spored agarics and boletes. The present paper contains descriptions of new agarics with rusty-brown or black spores discovered in the forested portion of eastern Texas.

The type collections have been deposited in the Herbarium of the University of Michigan, Ann Arbor, Michigan.

CONOCYBE ANTIPODA (Lasch) Kühner var. **humicola** var. nov.

Stipes non radicatus; sporae 7.8–10.9 × 4.5–5 × 6.2–7.8 μ; humi in nemoribus frondosis.

Pileus 0.9–1.3 cm broad, conic to subconic, unchanging or becoming convex with age, margin slightly incurved when young becoming straight when older, entire to eroded; surface moist, atomate, glabrous, striate to the disc when moist, evenly dark brown to umber ("snuff brown" [3] to "Saccardo's umber") to occasionally brown ("sayal brown") during all stages of development.

Flesh thin (c. 2 mm), brittle, concolorous with the surface; taste and odor not distinctive.

Lamellae close to subdistant, ascending-adnate to adnexed, becoming free with age, narrow, thin, light brown to pale fulvous ("clay color" to "ochraceous tawny") at maturity; margin entire, concolorous with the gill faces; lamellulae of 1–2 lengths.

Stipe 4–6.5 cm long, 0.5–1 mm broad at the apex, tapering slightly toward the apex, solid, translucent; surface glabrous except for white fibrils at the base, moist, olivaceous to yellow ("pale olive buff" to "Marguerite yellow") during all stages of development.

[1] Portion of a dissertation submitted to the Faculty of the University of Michigan in partial fulfillment of the requirements of the degree of Doctor of Philosophy.

[2] Grateful acknowledgment is made to Dr. A. H. Smith for his continued assistance and guidance in this study. Appreciation is also expressed to my wife, Ellen Thiers, for preparation of the Latin diagnoses.

[3] All colors given in quotations are from Ridgway, Color Standards and Nomenclature, Washington, D. C., 1912.

Spores bright rusty brown in KOH, flattened and obscurely angular in face view, ellipsoid in side view, germ pore terminal, 7.8–10.9 × 4.5–5 × 6.2–7.8 μ; pleurocystidia not differentiated; cheilocystidia abundant, hyaline, thin-walled, capitate, nine-pin-shaped, 14–17 × 5–8 μ; gill trama composed of hyaline, interwoven, filamentous hyphae c. 10 μ in diam.; cuticle differentiated as a palisade of hyaline, thin-walled, vesiculose to occasionally globose cells; pilocystidia and caulocystidia not differentiated; examined for clamp connections but none found.

Gregarious on plant debris in open oak woods. Open oak woods near clay pits, Wellborn, Brazos Co., Texas, May 18, 1953, *H. D. Thiers 1883*, TYPE.

This *Conocybe* is characterized by a rather dark pileus, fragile, buff to yellowish stipe and relatively small, flattened to obscurely angular spores. My collection differs from typical *Conocybe antipoda* in spore size, in habitat and in the apparent absence of a pseudorhiza. It appears to be closely related to *Conocybe teneroides*, which has smaller spores, very soft, fragile flesh, a tough stipe and a cinnamon brown rather than umber pileus.

Cortinarius acystidiosus sp. nov.

Pileus 4–5 cm latus, convexus demum plano-convexus, fibrillosus, viscidus, "light buff" demum "warm buff" vel "antimony yellow." Lamellae confertae vel subdistantes, adnatae vel adnexae, "pale pinkish cinnamon" demum "vinaceous buff" vel "avellaneous." Stipes 5–7 cm longus, 7–10 mm crassus, viscidus vel subviscidus, fibrillosus, albus, tactu "light buff." Sporae subglobosae vel ovoideae, 9.3–10.9 × 7.5–8 μ; cystidia nulla.

Pileus 4–5 cm broad, convex becoming plano-convex, margin strongly incurved when young, becoming slightly incurved to decurved with age, entire, sometimes with remnants of the cortina attached; surface sparsely covered with appressed, pale fulvous ("ochraceous tawny") fibrils, viscid, glistening; when young evenly colored yellowish ("light buff") changing to tan or yellow ("warm buff" to "antimony yellow") on the disc when older.

Flesh thick (4–6 mm), firm, white, unchanging; taste farinaceous, odor not distinctive.

Lamellae close to subdistant, adnate to broadly notched, pale brown ("pale pinkish cinnamon") when young becoming buff to lavender ("vinaceous buff" to "avellaneous") with age; margin pallid, entire to eroded; lamellulae of 2–3 lengths.

Stipe 5–7 cm long, 7–10 mm broad at the apex, equal except for clavate base, rooting, hollow; surface viscid to subviscid, appressed fibrillose, white, staining yellowish ("light buff") when bruised; cortina adhering to the stipe forming a fibrillose zone.

Spores rusty brown in KOH, subglobose to ovoid, roughened by a warty, wrinkled exosporium, thick-walled, 9.3–10.9 × 7.5–8 μ; basidia clavate, 4-spored, 20–25 × 7–10 μ; pleurocystidia and cheilocystidia not differentiated; gill trama composed of interwoven to subparallel, hyaline, filamentous hyphae c. 11 μ in diam.; pileus trama composed of compactly interwoven, hyaline hyphae c. 6 μ in diam.; cuticle composed of a brown (c. 75 μ) layer of loosely interwoven, hyaline, filamentous hyphae c. 4 μ in diam., partially gelatinizing in KOH; clamp connections present.

Scattered in humus in pine-hardwood forest. Sam Houston National Forest near Coldspring, San Jacinto Co., Texas, May 23, 1953, *H. D. Thiers 1902*, TYPE.

This *Cortinarius* is characterized by a viscid pileus and a slightly viscid, clavate stipe. It belongs in the subgenus *Myxacium*, and in the section *Clavati*. The absence of both pleurocystidia and cheilocystidia is of uncommon occurrence in this section. The spores, instead of being finely punctate, have a roughened, warty exosporium which appears loosely attached to the endosporium and occasionally breaks loose, reminding one of the spores of species of *Galerina*. The spore color in mass is darker than in most *Cortinarii*.

Cortinarius paliformis sp. nov.

Pileus 4–7 cm latus, late convexus, glaber, viscidus, "Verona brown" vel "cinnamon brown" demum centro "Mars brown" vel "russet." Lamellae adnatae, subdistantes, "cinnamon buff." Stipes 5–7 cm longus, 1–2 cm crassus, aequalis, viscidus, fibrillosus, "deep grayish lavender" vel "deep dull lavender." Sporae fusoideae vel subellipsoideae, 12.5–15.5 × 7.5–8.5 μ; pleurocystidia nulla; cheilocystidia vesiculosa.

Pileus 4–7 cm broad, broadly convex, margin with tints of lavender, decurved, entire; surface glabrous, very viscid, slightly rugose toward the margin; dark brown to red ("Mars brown" to "russet") on the disc, fading to brown ("Verona brown" to "cinnamon brown") on the margin.

Flesh firm, moderately thick (3–5 mm), concolorous with the surface; taste and odor not distinctive.

Lamellae adnate with a short decurrent tooth, subdistant, narrow, 3–4 mm thick, buff ("cinnamon buff") with definite lavender tints, margin eroded, slightly paler than the gill faces; lamellulae of three lengths.

Stipe 6 cm long, 1.5 cm broad at the apex, equal, solid, surface viscid, silky-appressed fibrillose, lavender ("deep grayish lavender" to "deep dull lavender"); fibrillose zone marking attachment of the cortina.

Spores bright rusty brown in KOH, roughened, fusoid to pip-shaped to subellipsoid, inequilateral, 12.5–15.5 × 7.5–8.5 μ; basidia clavate, 4-

spored, 43–47 × 10–12 μ; pleurocystidia not differentiated; cheilocystidia crowded, forming a sterile margin, saccate to vesiculose, hyaline, thin-walled, 23–31 × 19–23 μ; gill trama composed of large, irregularly shaped, thin-walled, hyaline cells; pileus trama composed of hyaline, interwoven, filamentous hyphae c. 4 μ in diam.; cuticle differentiated as a palisade of thin-walled, subgelatinous, hyaline, inflated cells with a turf of filamentous hyphal tips arising from them; clamp connections present.

Solitary in deep humus under loblolly pine in pine-hardwood forest. Pine-hardwood forest near Huntsville, Walker Co., Texas, April 14, 1953, *H. D. Thiers 1806*, TYPE.

The decidedly viscid pileus and stipe place this *Cortinarius* in the subgenus *Myxacium* and the *Splendidi* section. It seems to be rather closely related to the stirps *Elatior* but clamp connections are present. It has a type of cuticle that, as far as can be determined, is unique in the genus. Instead of having a typical gelatinous pellicle of narrow hyphae, it has a turf of relatively broad, hyaline hyphal tips which only partially gelatinize in KOH. It is further distinguished by the saccate to vesiculose cheilocystidia.

Inocybe texensis sp. nov.

Pileus 2–2.5 cm latus, subconicus demum convexus vel late conicus, siccus, fibrillosus, saepe squamosus vel rimosus, "clay color" vel "tawny olive" vel "sayal brown." Lamellae adnatae, 4–5 mm latae, confertae vel subdistantes, albae demum "snuff brown" vel "Saccardo's umber." Stipes 3–3.5 cm longus, 6–7.5 mm crassus, siccus, fibrillosus, rare fibrilloso-squamosus, "vinaceous." Sporae ellipsoideae vel angulatae vel verrucosae, 10.9–15.5 × 3.4–6.2 μ; pleurocystidia fusoideo-ventricosa vel cylindrica; cheilocystidia pleurocystidiis similia.

Pileus 2–2.5 cm broad, subconic becoming convex to broadly conic, margin slightly incurved when young, becoming straight and sometimes undulate and rimose with age; surface dry, appressed fibrillose, fibrils frequently forming suberect scales near the disc; colored more or less evenly brown ("clay color" to "tawny olive" to "sayal brown") during all stages of development.

Flesh thin (1–2 mm), concolorous with the surface; taste slightly bitter, odor not distinctive.

Lamellae ascending adnate, 4–5 mm broad, close to subdistant, narrow, thin, white when young, becoming brown ("snuff brown" to "Saccardo's umber") as the spores mature; margin entire, concolorous with the gill faces; lamellulae of three lengths.

Stipe 3–3.5 cm long, 6–7.5 mm broad at the apex, equal except for slight enlargement at the base, stuffed; surface dry, covered with short fibrils which are usually appressed to the surface but sometimes becoming

slightly recurved; lavender ("vinaceous") during all stages of development.

Spores rusty brown in KOH, smooth, ellipsoid to inequilateral to obscurely angular to nodulose with 1–3 nodules, 10.9–15.5 × 3.4–6.2 μ; basidia clavate, 4-spored, 30–34 × 6–8 μ; pleurocystidia abundant, hyaline, thin-walled, fusoid-ventricose to cylindric, usually having obtuse, incrusted apices, 45–54 × 12–23 μ; cheilocystidia abundant, similar to the pleurocystidia except smaller; gill trama subparallel, composed of hyaline, filamentous hyphae c. 8 μ in diam.; hymenium staining pale yellow in KOH; pileus trama interwoven, composed of hyaline, filamentous hyphae c. 8 μ in diam.; cuticle differentiated as a layer of radially arranged hyphae c. 7 μ in diam., staining dark brown in KOH; clamp connections present.

Solitary in sandy humus in open areas near loblolly pines. Sam Houston National Forest near Huntsville, Walker Co., Texas, April 19, 1952, *H. D. Thiers 1508*, TYPE.

The long, narrow, sparingly nodulose spores characterize this *Inocybe*. This character is supplemented by the fibrillose to subscaly surface, vinaceous, fibrillose stipe and thin-walled pleurocystidia. Stuntz (**4**) has described two other species with this type of spore. *Inocybe chelanensis* has a copious white veil. *Inocybe rainierensis* has a conspicuous, abrupt bulb at the base of the stipe and thick-walled cystidia.

Gymnopilus luteoviridis sp. nov.

Pileus 2.5–4 cm latus, convexus vel subconicus demum planus vel plano-convexus, siccus, fibrillosus demum fibrilloso-squamosus, "straw yellow" vel "mustard yellow," tactu viridis. Lamellae confertae vel subdistantes, adnatae vel adnexae, "cream buff" vel "chamois" demum "ochraceous tawny" vel "buckthorn brown." Stipes 4–6 cm longus, 3–5 mm crassus, siccus, glaber, "light buff" vel "warm buff," saepe tactu viridis. Sporae ellipsoideae vel subovoideae, 5.8–7 × 4.4–5.7 μ; pleurocystidia clavata vel fusoidea; cheilocystidia pleurocystidiis similia.

Pileus 2.5–4 cm broad, convex to subconic when young becoming plane to plano-convex, margin incurved when young, remaining decurved with age, entire to occasionally appendiculate when young, becoming eroded with age; surface dry, dull, conspicuously fibrillose, forming small recurved scales when older, scales pale fulvous ("ochraceous tawny") along the margin, changing to olivaceous ("dark olive") near the disc; more or less evenly colored dull yellow ("straw yellow") to "mustard yellow") to occasionally yellow ("primuline yellow"), entire carpophore staining green and drying with greenish tints.

Flesh moderately thick (4–5 mm), concolorcous with the surface; taste bitter, odor not distinctive.

Lamellae close to subdistant, adnate to adnexed with a short decurrent tooth, thin, 1–4 mm broad, buff to dark yellow ("cream buff" to "chamois") when young becoming tawny ("ochraceous tawny" to "buckthorn brown") with age; margin entire, concolorous with the gill faces; lamellulae of 1–2 lengths.

Stipe 4–6 cm long, 3–5 mm broad at the apex, tapering slightly toward the apex, stuffed to hollow; surface dry, glabrous, longitudinally striate, yellowish ("light buff" to "warm buff") during all stages of development, frequently staining greenish when old; annulus represented by a fibrillose zone near the apex.

Spores pale rusty brown in KOH, ellipsoid to subovoid, punctate-roughened, not truncate, thin-walled, 5.8–7 × 4.4–5.7 μ; basidia clavate, 4-spored, 18–25 × 6–8 μ; pleurocystidia abundant, clavate to fusoid, often contorted, staining bright ochraceous in KOH, 23–45 × 4–8 μ; cheilocystidia similar to pleurocystidia; gill trama composed of interwoven to parallel, filamentous hyphae c. 12 μ in diam.; cuticle differentiated as a broad (c. 75 μ) layer of incrusted, interwoven, filamentous hyphae c. 6 μ in diam., staining pale fulvous ("ochraceous tawny") in KOH; clamp connections present.

Gregarious to cespitose on oak stumps in dense hardwoods. Navasota River levee near College Station, Brazos Co., Texas, May 22, 1953, *H. D. Thiers 1894*, TYPE.

This fungus is distinguished by the distinctly yellow pileus, the greenish discolorations of the stipe and pileus and the presence of pleurocystidia. The green discolorations suggest a close relationship with *Gymnopilus aeruginosus* but the yellow pileus and the presence of pleurocystidia make it distinct.

GALERINA AUTUMNALIS (Peck) Kühner f. **robusta** f. nov.

Caespitosa; sporae in cumulo cinnamomeae, minute asperulae; basidia clavata, 14–17 × 6–9 μ.

Pileus 5–6.5 cm broad when fully expanded, obtusely convex when young, becoming plano-convex to plane when mature, colored ochraceous buff to yellow to pale fulvous ("antimony yellow" to "ochraceous tawny") in all stages of development; surface dry, glabrous; margin incurved when young, becoming decurved to plane with age, entire.

Lamellae adnate with a short decurrent tooth, close to subdistant, narrow, pale fulvous to brown ("ochraceous tawny" to "buckthorn brown"); margin entire, concolorous with the gill faces; lamellulae of three lengths.

Stipe 4–7.5 cm long, 4–8 mm thick at the apex, terete to flattened, cartilaginous, buff ("ochraceous buff") toward the apex and pale fulvous

("ochraceous tawny") toward the base, equal, hollow; surface dry, glabrous, longitudinally striate, basal portion usually covered with pallid hyphal tomentum; annulus represented by a fibrillose zone near the apex of the stipe.

Spores dull cinnamon in deposit, minutely roughened under oil except for plage, not truncate, apiculate, broadly ellipsoid to ovoid, 7.5–9.5 × 4.5–5.5 μ; basidia short-clavate, 4-spored, 14–17 × 6–9 μ; pleurocystidia scattered, elongate fusoid-ventricose and often with somewhat elongated apices, hyaline in KOH, 45–55 × 8–11 μ; cheilocystidia scattered to rare, similar to the pleurocystidia, no clavate to vesiculose cells present; gill trama parallel to subparallel, composed of large (c. 8 μ), filamentous hyphae; pileus trama interwoven, composed of large (c. 10 μ), filamentous hyphae; cuticle consisting of a narrow layer (c. 15 μ) of gelatinous, narrow (2.5–5 μ) hyphae; clamp connections present.

Densely cespitose on dead hardwood log in mixed pine-hardwood forest. Sam Houston National Forest near Richards, Montgomery Co., Texas, December 31, 1951, *H. D. Thiers 1432*, TYPE.

A number of minor characters, such as the small basidia, less roughened spores, and slightly more cinnamon color of the spore deposit in addition to the habit, combine to make this form distinct.

COPRINUS COMATUS (Fries) S. F. Gray var. **texensis** var. nov.

Stipes radicatus. Sporae 15.5–17.1 × 9.3–10.9 μ.

Pileus 2–5 cm high, 1–2 cm thick before expanding; narrowly ellipsoid to cylindric when young, expanding to plane or broadly umbonate, finally completely deliquescing, margin closely appressed to the stipe and attached to it by the partial veil when young, becoming strongly incurved and finally revolute before deliquescing, entire to eroded, sometimes appendiculate from partial veil remnants; surface dry, glabrous when young, cuticle becoming torn except at the disc as the cap expands, becoming fibrillose to fibrillose-scaly, tan to light brown ("clay color" to "cinnamon buff" or "ochraceous buff").

Flesh moderately thick (0.5–1 cm) toward the disc becoming very thin at the margin, white; taste and odor not distinctive.

Lamellae very crowded, moderately broad (c. 0.5 cm), free, white, eventually becoming black with mature spores and finally deliquescing; margin entire, concolorous with the gill faces.

Stipe 3–7 cm long, 7–17 mm broad at the apex, equal except for bulbous base, with a distinct pseudorhiza, hollow, usually having superior, white, fibrillose annulus which becomes movable with age; surface white, dry, obscurely fibrillose, frequently becoming glabrous, longitudinally striate.

Spores black in mass, dark brown in KOH, smooth, broadly ellipsoid to ovoid, sometimes obscurely angular, germ pore apical or very slightly eccentric, 15.5–17.1 × 9.3–10.9 μ; basidia clavate, trimorphic, 4-spored, 35–42 × 13–21 to 17–24 × 9–14 μ; pleurocystidia not differentiated; paraphyses scattered, nearly globose to saccate, hyaline, thin-walled, 28–35 × 17–23 μ; cheilocystidia numerous, vesiculose to saccate, hyaline, thin-walled, 47–60 × 19–28 μ; gill trama interwoven, composed of hyaline, filamentous hyphae c. 6 μ in diam.; pileus trama interwoven, hyaline, composed of filamentous hyphae c. 6 μ in diam.; cuticle composed of a layer of radially arranged, filamentous hyphae c. 10 μ in diam.; examined for clamp connections but none found.

Gregarious in humus under hardwood trees in mixed forest. Sam Houston National Forest near the San Jacinto River, Montgomery Co., Texas, February 23, 1952, *H. D. Thiers 1469*, TYPE.

This very distinctive species is easily recognized by the long cylindrical pileus, and the cuticle which breaks up into numerous conspicuous scales at maturity. This variety is distinct from the typical variety by having large spores and a distinct, well-developed pseudorhiza.

Coprinus subcurtus sp. nov.

Pileus 0.3–0.5 cm longus, 0.2–4 mm latus, conicus vel cylindricus demum planus vel inaequalis, siccus, granulosus, centro "sayal brown" vel "ochraceous tawny," margine "pale smoke gray" vel "pale olive gray." Lamellae confertissimae, adnatae demum liberae, angustae. Stipes 2–2.5 cm longus, 0.5–1 mm crassus, aequalis, albus, siccus, glaber. Sporae ellipsoideae, angulatae, 10.4–11 × 9.5–10 × 6–6.8 μ; pleurocystidia nulla; cheilocystidia globosa vel vesiculosa.

Pileus 0.3–0.5 cm high and 0.2–0.4 mm across before expansion, conic to cylindrical becoming plane to flared and eventually revolute before deliquescence, margin entire to irregularly lobed or incised, closely appressed to the stipe at first, becoming straight to flared with age; surface dry, covered with reddish tan ("ochraceous tawny") granules on the disc that become concolorous with the surface at the margin, brown ("sayal brown" to "ochraceous tawny") on the disc changing to light gray ("pale smoke gray" to "pale olive gray") toward the margin.

Flesh very thin (0.5–1 mm), concolorous with the surface, deliquescing; taste and odor not distinctive.

Lamellae crowded, ascending adnate becoming free with age, narrow, thin, white when young becoming black as the spores mature, deliquescing; margin entire, concolorous with the gill faces; lamellulae absent.

Stipe 2–2.5 cm long, 0.5–1 mm broad at the apex, solid to stuffed, equal to slightly bulbous at the base with white, coarse mycelial hairs attached, white during all stages of development; surface dry, glabrous.

Spores black in mass, dark brown in KOH, ellipsoid in side view, angular in face view, flattened, germ pore apical, 10.4–11 × 9.5–10 × 6–6.8 μ; basidia dimorphic, 4-spored, clavate to subcylindric, 18–23 × 6–9 and 9–12 × 5–8 μ; pleurocystidia not differentiated; cheilocystidia scattered, saccate to vesiculose, hyaline, thin-walled, 26–40 × 26–31 μ; gill trama composed of a narrow layer of interwoven, filamentous hyphae c. 4 μ in diam., staining yellowish ("light buff") in KOH; pileus trama scant, composed of hyaline, interwoven, filamentous hyphae c. 5 μ in diam.; cuticle composed of a layer of loosely attached, spherical, hyaline, thin-walled cells of variable size; examined for clamp connections but none found.

Gregarious on old cow dung in open oak woods. Open oak woods near Millican, Brazos Co., Texas, May 16, 1953, *H. D. Thiers 1844*, TYPE.

The nature of the universal veil places this *Coprinus* in the section *Farinosi*. This fungus does not satisfactorily agree with any of the published descriptions in that section. The presence of cheilocystidia, absence of pilocystidia and larger spores distinguish it from *Coprinus curtus*, to which it appears closely related.

Coprinus subangularis sp. nov.

Pileus 0.6–1.3 cm longus, 3–7 mm latus, conicus demum convexus, fibrillosus demum glaber, siccus, albus vel "cartridge buff." Lamellae liberae, confertissimae, angustae. Stipes 1–2 cm longus, 1–2 mm crassus, aequalis, albus, siccus, fibrillosus. Sporae angulatae, 6–8 × 3–3.5 μ; pleurocystidia fusoideo-ventricosa vel subclavata; cheilocystidia globosa vel vesiculosa.

Pileus 0.6–1.3 cm high and 3–7 mm across before expanding, conic at first, becoming campanulate to convex and finally revolute before deliquescing, margin closely appressed to the stipe, white-fimbriate when young, becoming glabrous with age, entire; surface densely covered with loosely attached fibrils, usually glabrescent before expanding, fibrils frequently becoming recurved, giving a squarrose appearance; white to pale ivory ("cartridge buff") when young, becoming various shades of gray as the spores mature.

Flesh very thin (0.5–1 mm), concolorous with the surface, deliquescing; taste and odor not distinctive.

Lamellae free, crowded, thin, narrow, white when young, becoming progressively darker as the spores mature, deliquescing; margin entire, concolorous with the gill faces; no lamellulae present.

Stipe 1–2 cm long, 1–2 mm broad at the apex, equal, solid to stuffed, white during all stages of development; surface dry, covered with white, appressed fibrils.

Spores black in mass, black to dark brown in KOH, obscurely angular, smooth, moderately thick-walled, 6–8 × 3–3.5 μ; basidia dimorphic, clavate, 4-spored, 15–17 × 8–10 μ; pleurocystidia scattered, fusoid-ventricose to subclavate, hyaline, thin-walled, 15–19 × 14–19 μ; cheilocystidia numerous, hyaline, saccate to vesiculose, thin-walled, 15–25 × 14–23 μ; gill trama scant, composed of interwoven, filamentous hyphae c. 6 μ in diam.; pileus trama thin, composed of interwoven, hyaline filamentous hyphae c. 5 μ in diam.; cuticle differentiated as a palisade of vesiculose to clavate cells, hyaline, thin-walled; examined for clamp connections but none found.

Gregarious on old cow dung in pine-hardwood forest. Sam Houston National Forest near Richards, Montgomery Co., Texas, May 16, 1953, *H. D. Thiers 1845*, TYPE.

The obscurely angular and flattened spores, differently shaped cystidia and a different substrate separate this species from *Coprinus pseudoradiatus*, to which it appears most closely related.

AGARICUS PLACOMYCES Peck var. **flavescens** var. nov.

Pileus margine "ivory yellow," in sicco "ochraceous buff."

Pileus 8–12 cm broad, convex when young, becoming broadly convex to obscurely broadly umbonate with age, margin incurved at first, becoming plane to broadly decurved with age, extending 1–2 mm beyond the lamellae, entire to eroded; surface dry, covered with numerous, loosely appressed to slightly recurved, fibrillose scales, umber to lavender ("Saccardo's umber" to "fuscous" to "wood brown"), areas between the scales umber to gray ("Saccardo's umber" to "light drab") toward the disc and ivory ("ivory yellow") toward the margin, drying reddish tan ("ochraceous buff").

Flesh moderately thick (4–5 mm), white to pale tan ("pale ochraceous buff"), unchanging or slowly changing to pinkish tan ("pale pinkish buff") when bruised; taste and odor mild.

Lamellae free, close to crowded, narrow, thin, white when young becoming purple ("light russet vinaceous" to "russet vinaceous") and finally nearly black; margin entire to obscurely serrate, white to concolorous with the gill faces; lamellulae of 2–3 lengths.

Stipe 7–13 cm long, 1–2 cm broad at the apex, tapering slightly toward the apex, base slightly enlarged and often with rhizoids attached; surface dry, glabrous, white when young, becoming purple ("russet vinaceous") above the annulus and lavender to olivaceous ("vinaceous buff" to "drab") toward the base when older, staining dark yellow ("chamois") when bruised, occasionally unchanging when bruised; annulus ample, pendant, superior, attached, glabrous above with soft, floccose patches below, whitish to pale fulvous ("ochraceous tawny").

Spores dark chocolate brown in KOH, ellipsoid to oblong to subovoid, smooth, thin-walled, 4.5–6.2 × 2.8–3.5 μ; basidia clavate, 4-spored, 15–20 × 6–8 μ; pleurocystidia not differentiated; cheilocystidia scattered to numerous, hyaline, thin-walled, globose to clavate, 12–16 × 6–9 μ; gill trama subparallel to slightly interwoven, composed of hyaline, filamentous hyphae c. 8 μ in diam.; pileus trama composed of large (c. 13 μ), hyaline, loosely interwoven, filamentous hyphae; cuticle differentiated as a narrow (c. 20 μ) layer of radially arranged hyphae, c. 8 μ in diam., usually staining ochraceous in KOH; examined for clamp connections but none found.

Solitary in humus in hardwood or pine-hardwood forests. Hardwoods near clay pits, Wellborn, Brazos Co., Texas, June 7, 1952, *H. D. Thiers 1656*, TYPE. Sam Houston National Forest near Richards, Montgomery Co., Texas, May 18, 1953, *H. D. Thiers 1895*.

This fungus is closely related to *Agaricus silvaticus* and is distinguished from it by the dark fibrils on the surface of the pileus. Variety *flavescens* differs from the typical *Agaricus placomyces* in that the surface of the pileus becomes yellow on the disc when old, and dries ochraceous to yellow. The typical variety has not as yet been found in this state.

PSATHYRELLA HYMENOCEPHALA (Peck) A. H. Smith var. **farinacea** var. nov.

Pileus centro "bister," margine "snuff brown" vel "tawny olive" demum rare "warm buff" vel "mummy brown." Sapor farinarius.

Pileus 1.5–3 cm broad, conic to convex becoming subconic to campanulate, margin incurved when young, becoming straight to flared when older, appendiculate with white veil elements when young, becoming glabrous with age; surface moist, glabrous, frequently becoming striate when older, when young dark brown ("bister") on the disc, changing to brown or olivaceous ("snuff brown" to "tawny olive") toward the margin, when older sometimes fading to yellowish ("warm buff") or darkening to dark brown ("mummy brown").

Flesh thin (1–2 mm), brittle, concolorous with the surface; taste farinaceous, odor not distinctive.

Lamellae ascending-adnate, close to subdistant, narrow, thin, yellowish ("light buff") when young, becoming lavender ("avellaneous" to "wood brown") when older; margin entire, concolorous with the gill faces; no lamellulae present.

Stipe 5–8 cm long, 2.5–4 mm broad at the apex, tapering slightly toward the apex, stuffed to hollow; surface dry, glabrous when young, frequently becoming fibrillose when older; white during all stages of development.

Spores smoky umber in KOH, oblong to subovoid, germ pore terminal, occasionally narrowed in the middle, smooth, thin-walled, 5.8–7.5 × 3.1–4.2 μ; basidia clavate, 4-spored, 13–17 × 8–10 μ; pleurocystidia not differentiated; cheilocystidia abundant, clavate to cylindric to obscurely fusoid-ventricose, hyaline, thin-walled, 33–50 × 11–16 μ; gill trama scant, hyaline, interwoven, composed of filamentous hyphae c. 5 μ in diam.; pileus trama scant, composed of interwoven, filamentous hyphae c. 9 μ in diam., staining brown ("buckthorn brown") in KOH; cuticle differentiated as a palisade of thin-walled, vesiculose to clavate, hyaline cells; clamp connections present.

Gregarious to scattered in humus in the vicinity of decaying hardwood logs. Sam Houston National Forest near Richards, Montgomery Co., Texas, May 17, 1953, *H. D. Thiers 1866*, TYPE.

This variety is distinguished by the farinaceous taste. Smith (**3**) has made a thorough study of this species. It belongs in the same complex as *Psathyrella candolleana* and *Psathyrella coronata*. The smaller pileus and dark brown color which fades to almost white with age make it distinct. The rapid color change observed by Kauffman (**1**) and Parker (**2**) for *Psathyrella saccharinophilum* and *Psathyrella cinereum*, which Smith reduces to synonymy with *Psathyrella hymenocephala*, was not observed in my material.

DEPARTMENT OF BIOLOGY
SAN FRANCISCO STATE COLLEGE
SAN FRANCISCO, CALIFORNIA

LITERATURE CITED

1. **Kauffman, C. H.** The Agaricaceae of Michigan. Mich. Geol. and Biol. Surv. Pub. *26*, Biol. Ser. No. 5, Vols. I–II. 1918.
2. **Parker, C. S.** A taxonomic study of the genus *Hypholoma* in North America. Mycologia **25**: 160–212. 1933.
3. **Smith, A. H.** Unusual agarics from Michigan. II. Pap. Mich. Acad. Sci. Arts Letters **30**: 171–183. "1934" (1935).
4. **Smith, A. H., and D. E. Stuntz.** New or noteworthy fungi from Mt. Rainier National Park. Mycologia **42**: 80–134. 1950.
5. **Thiers, H. D.** The agaric flora of Texas. I. New species of agarics and boletes. Mycologia **49**: 707–722. 1957.
6. **Thiers, H. D.** The agaric flora of Texas. II. New species of white- and pink-spored agarics. Mycologia **50**: 514–523. 1958.

STUDIES OF SOME TREMELLACEAE. III. THE GENUS BOURDOTIA [1]

KENNETH WELLS [2]

(WITH 9 FIGURES)

The taxon *Bourdotia* was originally described by Bresadola (**1908**) as a subgenus of *Sebacina* Tul. to include a single species, *S. galzinii* Bres. The distinguishing character was the possession of gloeocystidia in the hymenium. Bresadola and Torrend (in Torrend, **1913**) later elevated *Bourdotia* to the rank of genus and described an additional species.[3] Later authors have accorded the group varied treatment. Rea (**1922**), Gäumann and Dodge (**1928**), and Killermann (**1928**) recognized *Bourdotia* as a subgenus; however, Killermann included one *Sebacina*-like species with gloeocystidia in the genus *Exidiopsis* (Bref.) Möll. and another gloeocystidiate species in an unnamed group consisting of an assortment of species now usually segregated into several genera. Burt (**1915, 1926**) included the species of the *Bourdotia* group in the genus *Sebacina*. Bourdot and Galzin (**1928**) adopted Bresadola and Torrend's concept of the group and recognized five species as well known and three species as insufficiently known. Neuhoff (**1936**) followed Bourdot and Galzin in his treatment of the Swedish species. More recently Malençon (**1954**) in his treatment of certain species from the Atlas Mountains of North Africa separated *Bourdotia* as a genus. Rogers (**1933, 1935**) recognized the gloeocystidiate species of *Sebacina*

[1] Based on a portion of a thesis submitted to the Graduate College of the State University of Iowa in partial fulfillment of the requirements for the degree of Doctor of Philosophy, under the direction of Professor G. W. Martin.

[2] Present address: Department of Botany, University of California, Davis, California.

[3] Donk (Taxon **7**: 173. 1958) has recently criticized the recognition of Bresadola and Torrend as authors of the generic name, preferring to cite Trotter (in Saccardo, Syll. Fung. **23**: 571. 1925) since the former authors did not supply a generic description; however, it is, in my opinion, preferable to follow Martin (personal communication) and accept Bresodola and Torrend as having validly published the generic name in 1913. There should be little cause for confusion in interpreting the generic concept of these authors, since it was clearly their intent to elevate the name to generic status in their 1913 publication and since they refer to Bresadola's earlier paper in which a subgeneric description is given.

as a subgenus, concluding that *Bourdotia* is a homogeneous group since the species studied possess gloeocystidia, have similar basidial structure, and possess a basal layer of agglutinate hyphae from which arise the gloeocystidia and fertile hyphae, and the fructifications have similar texture.

In the most complete treatment to date, McGuire (**1941**) segregated *Bourdotia* as a section of *Sebacina*. McGuire suggested that the arid species form a continuous series which may possibly have evolved from an arid, non-gloeocystidiate species, perhaps *Sebacina calcea* (Pers. ex Fries) Bres. McGuire did not consider the group as a homogeneous segregate, suggesting that the waxy-gelatinous species, such as *B. galzinii*, have a separate origin. Martin (**1952**) followed McGuire's classification of the genus *Sebacina*, indicating, however, that he regarded the group as a heterogeneous assemblage of species.

Olive (**1958**) in his treatment of the Tremellales of Tahiti criticized the recognition of *Bourdotia* as a genus solely on the basis of the presence of gloeocystidia. In the specimens which have been available for study, I have found that the three main characters used by Rogers (**1933**) to define *Bourdotia* as a subgenus and by Ervin (**1957**) to segregate the group as a genus are constant and recognizable in all but very immature or overly mature specimens. First, all specimens have gloeocystidia which arise from a horizontal hyphal layer and which develop a yellow-granular content at maturity. The two known collections of *Sebacina obscura* do not have a granular content within the gloeocystidia; however, it is conceivable that both collections were not mature. The gloeocystidia do not usually project above the hymenium; however, in the simpler species or in young specimens a well-defined hymenium is often absent and the gloeocystidia may then project above adjacent basidia. In the genus *Exidiopsis* cylindrical or inflated hyphal elements, which do not usually project above the hymenium, are present in some species (e.g., *Exidiopsis fuliginea* Rick; see Wells, **1957**), but such elements either arise from the fertile hyphae or from the same hyphae as the slender branching dikaryophyses and lack the yellow-granular contents of the gloeocystidia of the genus *Bourdotia*. As noted by Olive (**1958**) in *Exidiopsis mucedinea* (Pat.) Wells and by Wittlake (**1938**) in *Sebacina calcea*, such hymenial elements frequently intergrade with the slender branching dikaryophyses. These structures are not, in my opinion, homologous with the yellow-granular gloeocystidia of the genus *Bourdotia*.

Second, the fertile hypha is generally an upright strand bearing a cluster of turgid basidia at the apex and, in older specimens of most

species, with the collapsed basidia forming an involucre-like sheath around the fertile hyphal strand and the turgid basidia. This type of basidial development is not strikingly apparent in such species as *B. galzinii* and *B. petiolata*, since the basidia are formed at greater intervals along the fertile hyphae; however, the same method of development is present in these species as in the more arid species. This character is most conspicuous in such species as *B. grandinioides*, *B. eyrei*, *B. caesio-cinerea*, *Sebacina pini*, *B. cinerea*, and *Sebacina spinosa*. In such species as *Heterochaete burtii*, *Sebacina obscura*, and *Sebacina aspera* the collections are as yet too few to reveal the possible variations within the species; however, the method of basidial development would indicate that such an involucre-like sheath would be formed in more mature fructifications. Certainly the important character is the method of basidial formation which leads to the more conspicuous character of a sheath of collapsed basidia, and not that the collapsed basidia surrounding the fertile hyphae are present in all collections.

A third distinguishing feature of this genus is the structure of the fructification. There is present a basal layer of agglutinate hyphae parallel with the substrate from which the gloeocystidia and fertile hyphae arise. This basal layer may consist of only a few strands, as in *Sebacina obscura*, or it may form a considerable portion of the fructification, as in *Heterochaete burtii*. It has also been noted that if two or more growth layers are present, each layer is subtended by a horizontal hyphal layer from which the fertile hyphae and most of the gloeocystidia arise. In some cases the gloeocystidia may extend through two or more growth layers. Tropical collections of *B. cinerea* have been examined with six to eight such layers.

The basidia are quite varied in form. In *Sebacina obscura* the basidia are globose with truncate apices and lack differentiated epibasidia. In *B. grandinioides*, *B. eyrei*, *B. caesio-cinerea*, and *Sebacina spinosa* epibasidia are not clearly apparent and the form of the basidia varies from subglobose to suburniform. In *Sebacina pini* and *S. aspera* epibasidia are not a conspicuous part of the basidium, which varies in form from subglobose to suburniform. In *B. cinerea* the basidia are generally obovate to elongate with distinguishable epibasidia. *B. petiolata* and *Heterochaete burtii* have clavate basidia with apparent epibasidia. *B. galzinii* possesses basidia which have well-developed epibasidia and are ovate, obovate, or pyriform in shape. The basidia of *B. galzinii* are quite similar to the basidia found in the genus *Ductifera* with respect to form, structure, and manner of septation.

The basidiospores within the genus vary from the elliptical-fusoid spores of *Sebacina obscura* to the near-globose spores of *B. grandinioides*, *B. eyrei*, and *B. caesio-cinerea*. The spores of *B. cinerea* and *B. petiolata* vary from subglobose to cylindrical. In *B. galzinii* and *Sebacina pini* the spores are generally cylindrical. *Heterochaete burtii* is the only species included in this genus in which the basidiospores have been found to be consistently cylindrical-curved. Olive (**1958**) has recently described two species, *Sebacina spinosa* and *S. aspera*, from Tahiti which obviously belong to this genus, with unique warty or spiny spores. The spores of *B. caesio-cinerea* are often minutely spinose.

Proceeding from the species which is simplest in structure, *Sebacina obscura*, to the most complex species, *B. galzinii*, it is possible to observe rather consistent modifications of several characters, such as the form of the basidia, including the protuberances upon which the basidiospores are formed, intervals between the basidia, form of the basidiospores, dikaryophyses, and texture of the fructification. Therefore it does not appear desirable to postulate a dual origin for the *Bourdotia* group. *Sebacina obscura* is the most primitive species, if simplicity is an indication of primitiveness; however, it is, of course, possible that the entire group could have arisen by reduction from *B. galzinii*.

There is a possible close relation of *Heterochaetella dubia* (Bourd. & Galz.) Bourd. & Galz. and *Sebacina minima* Olive with *Bourdotia*, as suggested by Olive (**1958**); however, the exact basidial structure of *H. dubia* has not, to my knowledge, been described. This should be accomplished before any generic transfers are made.

Portions of all specimens examined are deposited in the Mycological Herbarium of the State University of Iowa or in the personal herbarium of Dr. D. P. Rogers, unless indicated otherwise by the use of the herbarium symbols recommended by Lanjouw and Stafleu (**1956**).

I am greatly indebted to Dr. G. W. Martin for his helpful criticism and advice. Acknowledgments are due Dr. D. P. Rogers for his clarification of certain nomenclatural problems and for the loan of numerous specimens, to Dr. Clark T. Rogerson for the loan of several specimens from the New York Botanical Garden, to Dr. I. M. Lamb for the loan of specimens from the Farlow Herbarium, to the Director, Royal Botanic Gardens, Kew, for the loan of type specimens of *Sebacina eyrei* Wakef., and especially to Prof. Roger Heim for the loan of a number of specimens from the Herbarium of H. Bourdot.

DESCRIPTION OF GENUS

BOURDOTIA (Bres.) Bres. & Torrend, in Torrend, Brotéria sér. bot. **11**: 88. 1913.

Sebacina subg. *Bourdotia* Bres., Ann. Myc. **6**: 46. 1908.

Basidiodendron Rick, Brotéria **25**: 74. 1938.

Resupinate; margins indeterminate and adnate; texture waxy-gelatinous, arid-waxy, to arid; fructification consisting of a horizontal layer of indistinct hyphae from which arise the gloeocystidia, fertile hyphae, and, if present, dikaryophyses; gloeocystidia becoming yellowish- to brownish-granular; fertile hyphae forming basidia in acropetal succession, collapsed basidia usually persisting to form an involucre-like sheath along the fertile hyphae; basidia various in shape, becoming longitudinally septate into 2–4-cells; epibasidia tubular, attenuate, or lacking; basidiospores germinating by repetition or by the formation of germination tube.

TYPE: *Sebacina* (*Bourdotia*) *galzinii* Bres.

KEY TO SPECIES

a. Fructification waxy-gelatinous; dikaryophyses much-branched and forming a definite layer above the basidia.....b
a. Fructification waxy, arid-waxy, to arid; dikaryophyses, if present, simple to little-branched.....c
b. Basidia obovate to ovate, not becoming separated into stalk and fertile portion by diverging longitudinal septa; basidiospores cylindrical to ovate, 8–13.5 × 4.5–7.5 μ.....1. *B. galzinii*
b. Basidia clavate, becoming delimited into basal stalk and apical fertile portion by diverging longitudinal septa; spores oblong, ellipsoidal to subglobose, 7–11 × 6–8 μ.....2. *B. petiolata*
c. Cone-shaped mineral accumulations piercing the hymenium forming macroscopically visible tubercles; basidiospores cylindrical-curved, 9–12 × 4–5 μ.....3. *B. burtii*
c. Cone-shaped mineral accumulations not present; spores cylindrical, elliptical-fusoid, elliptical to subglobose.....d
d. Basidiospores mostly 8 μ or larger in diameter; basidia 14 μ or larger in diameter.....e
d. Basidiospores and basidia smaller.....f
e. Basidiospores becoming thick-walled and spiny or warty, 12.5–18 μ in diameter.....4. *B. aspera*
e. Basidiospores remaining relatively thin-walled and smooth, ovate to cylindrical, 12–22 × 8–12 μ, with 1 to several large, conspicuous oil globules..5. *B. pini*
f. Basidia usually obovate to elongate; basidiospores oblong to broadly ovate, less often subglobose, 7–12 × 4–10 μ; definite epibasidia present.....6. *B. cinerea*
f. Basidia subglobose to urniform, basidiospores elliptical-fusoid or elliptical to subglobose; epibasidia lacking or subulate and not clearly differentiated.....g

g. Basidiospores subglobose, with symmetrically attached prominent apiculus, sometimes with spinose walls.....h
g. Basidiospores elliptical to subglobose, with minute apiculus or with rather prominent apiculus obliquely attached.....i
h. Spores thin-walled, smooth to minutely spinose, 6–11 μ in diameter.....7. *B. caesio-cinerea*
h. Spores thick-walled, conspicuously spinose, 5–8 μ in diameter....8. *B. spinosa*
i. Surface porous-reticulate to continuous; mature basidia usually urniform, 8.5–12.5 × 6–8.5 μ; basidiospores subglobose to elongate, 4.5–7 × 4–6 μ.....9. *B. eyrei*
i. Surface pruinose, arachnoid, or porous-reticulate, sometimes with fertile columns; mature basidia subglobose to suburniform; basidiospores subglobose, elliptical, or elliptical-fusoid, if subglobose then 3–5 μ in diameter...j
j. Basidiospores elliptical to subglobose with short axis through apiculus, 3–5 × 3–4 μ; surface pruinose to porous-reticulate and developing scattered obtuse columns; gloeocystidia becoming yellow-granular.....10. *B. grandinioides*
j. Basidiospores elliptical-fusoid with long axis through apiculus, 9–10 × 3.5–4.5 μ; surface arachnoid; gloeocystidia remaining hyaline in known specimens.....11. *B. obscura*

ACCEPTED SPECIES

1. BOURDOTIA GALZINII (Bres.) Bres. & Torr., in Torrend, Brotéria sér. bot. **11**: 88. 1913. FIG. 1

Sebacina (*Bourdotia*) *galzinii* Bres., Ann. Myc. **6**: 46. 1908.

Bourdotia caesia Bres. & Torr., in Torrend, Brotéria sér. bot. **11**: 88. 1913.

Bourdotia pululahuana (Pat.) Bourd. & Galz. ssp. *galzinii* (Bres.) Bourd. & Galz., Hymén. Fr. 48. 1928. Not *Tremella pululahuana* Pat.

Bourdotia pululahuana (Pat.) Bourd. & Galz. ssp. *caesia* (Bres. & Torr.) Bourd. & Galz., Hymén. Fr. 48. 1928.

Sebacina pululahuana sensu Rogers, Univ. Iowa Stud. Nat. Hist. **17**: 38. 1935, pro parte. Not *Tremella pululahuana* Pat.[4]

Soft to waxy-gelatinous, effused, hyaline to grayish-hyaline; surface smooth to undulate; margins adnate and indeterminate; drying to an ochraceous or dark vernicose layer; often with scattered mineral accretions embedded in the fructification and becoming prominent upon dry-

[4] Rogers cites four specimens in his description, all of which I have been able to examine. *G. W. Martin 1398* and *1399* and Herb. M. A. Donk 5707 should, in my opinion, be included in *B. galzinii*. As previously reported (Wells, **1957**, **1958**), the type of *Sebacina lactescens* Burt is best included within the concept of *Ductifera sucina* (Möll.) Wells. The combination *Sebacina pululahuana* (Pat.) Rogers is to be regarded as a later synonym of *Tremella pululahuana* Pat.

ing; fructification (25–)50–300 μ thick, consisting of a basal hyphal layer of indistinct, loosely arranged hyphae, parallel with the substrate, from which the gloeocystidia, dikaryophyses, and fertile hyphae arise; often consisting of several growth layers each composed of a horizontal hyphal layer giving rise to a hymenial layer; gloeocystidia at first hyaline, becoming yellowish- or brownish-granular and flexuous, cylindrical, fusiform, or clavate, sometimes expanded to form a globose apex, often extending through several growth layers, (25–)35–170(–270) × 2.5–7.5(–13) μ; dikaryophyses finely-branched, somewhat nodulose, becoming indistinct, 1–2 μ in diameter and arising from the fertile hyphae, subclavate and terminated by fine branches, dikaryophyses forming a layer 10–20 μ above the basidia; fertile hyphae 1.5–3 μ in diameter, forming basidia in an acropetal series by proliferation through clamp-connections at the base of the basidia; basidia arising as clavate structures, becoming ovate, obovate to pyriform and 4-celled by longitudinal division, the collapsed basidia in older spicemens persisting as a loose involucre-like sheath along the fertile hyphae, guttulate, 12–20(–24) × (7.5–)9–13 μ; epibasidia tubular, up to 20 μ in length, 2.5–3 μ in diameter, basidiospores cylindrical to ovate, laterally depressed or slightly curved, 8–13.5 × 4.5–7.5 μ, germinating by repetition.

On dead wood. Known from Ontario, New England, Iowa, Panamá, West Indies, Europe, and North Africa.

Type locality: France.

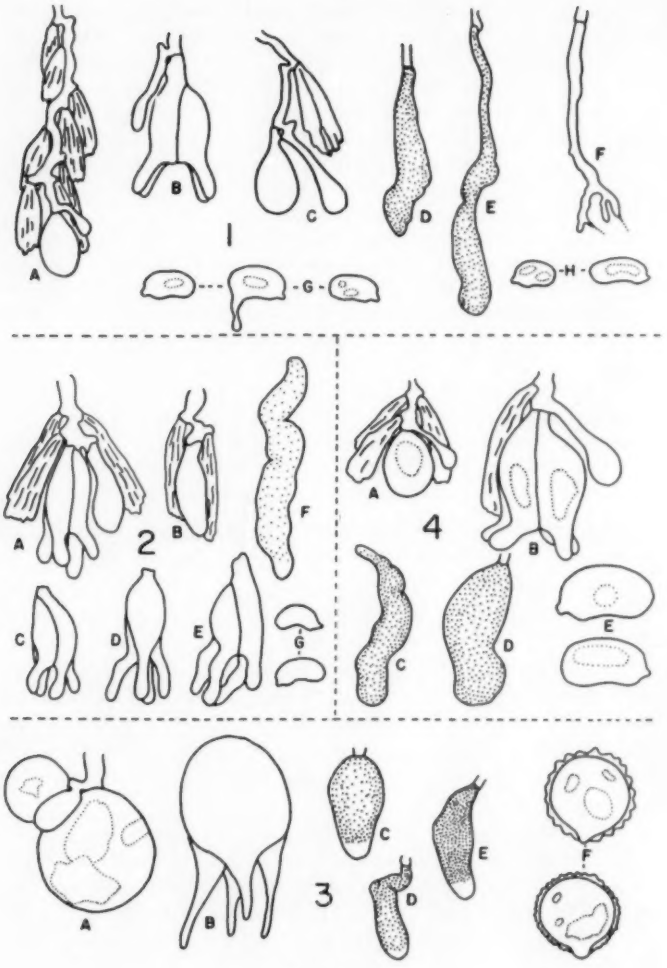
Illustrations: Brotéria sér. bot. **11**: 88, *f. 9;* Bourd. & Galz., Hymén. Fr. 48, *f. 25;* Univ. Iowa Stud. Nat. Hist. **17**: 35, *pl. 3, f. 17;* **19**: *pl. 1, f. 10;* Lloydia **4**: 34, *f. 80–82;* Bull. Soc. Myc. Fr. **70**: 123, *f. 1, f.*

Specimens examined: North America: Ontario, TRT 16833, 16841; Maine, *G. W. Martin 5058;* Massachusetts, *A. M. & D. P. Rogers, 1010, 2275;* Iowa, several collections, including *GWM 1380, 1398, 1399.* Panamá: Prov. Chiriquí, *GWM 2451.* Austria: Herb. M. A. Donk 5707.

A well-defined species, easily distinguished because of the gelatinous texture, the finely-branched dikaryophyses forming a definite layer above the basidia, the usually obovate to pyriform basidia, and the near-cylindrical basidiospores. It is easily separated from the genus *Ductifera* by the lack of an ascending hyphal layer and the completely effused growth habit.

2. BOURDOTIA PETIOLATA (Rogers) Wells, Lloydia **20**: 54. 1957.
Sebacina petiolata Rogers, Pacific Science **1**: 99. 1947.

This species was described and illustrated in Lloydia **20**: 54, *f. 7.* 1957. A collection by Dr. A. L. Welden (*Welden 488*) extends the



FIGS. 1–4.

FIG. 1. *Bourdotia galzinii.* A–C. Portions of fertile hyphae, × 875 (A and B from *G. W. Martin 5058;* C from *GWM 1399*). D, E. Glococystidia, × 875 (D from *GWM 2451;* E from *GWM 1399*). F. Dikaryophysis, × 875 (*GWM*, 5 Sept., 1943). G. Basidiospores, × 875 (left and center spore from *GWM 1399;* right spore from *GWM 2451*). H. Basidiospores, × 875 (left from *GWM 2451;*

known range to Jamaica. This collection is similar to the specimens previously examined except that the gloeocystidia were somewhat longer, up to 160 μ in length, and the basidiospores slightly larger. Olive (**1958**) has reported this species from Tahiti.

3. **Bourdotia burtii** (Bres.) comb. nov. FIG. 2

Heterochaete burtii Bres., Ann. Myc. **18**: 51. 1920.

Arid-waxy, effused, indeterminate, ochraceous to grayish pruinose; surface strongly tuberculate, tubercles usually nearly white and abundant; margins often somewhat lighter and adnate; drying to an ochraceous or gray crust; fructification 50–220 μ in thickness, consisting of a basal layer of agglutinate hyphae 2.5–4 μ in diameter, arranged parallel with the substrate, becoming light ochraceous in older specimens, sometimes arranged in strands and forming a loose network surrounding large, mineral accretions, basal hyphal layer giving rise to the gloeocystidia, dikaryophyses, and fertile hyphae; in some areas large cone-shaped mineral accretions are present over which the fructification is formed, resulting in a macroscopically visible tubercle; gloeocystidia cylindrical, subclavate, or subfusiform, at first hyaline, tardily becoming flexuous and light yellow-granular, sparse to abundant, 30–70 × (2–)3.5–9 μ; dikaryophyses sparse, soon becoming indistinct, simple to little-branched, 1.5–3 μ in diameter; fertile hyphae fluexuous, 2–4(–5) μ in diameter, forming basidia in a close acropetal series; probasidia arising as clavate structures subtended by clamp-connections, becoming elongate to obovate and 2–4-celled, (13.5–)15–20.5(–25) × 7–9 μ, sometimes guttulate; epibasidia tubular, up to 15 μ in length, 3–4 μ in diameter; basidiospores cylindrical-curved, 9–12 × 4–5 μ.

On dead wood, often corticated. Known from Louisiana, Panamá, and Brazil.

Type locality: St. Martinsville, Louisiana.

Specimens examined: Panamá: Canal Zone, Valley of Rio Chagres, *G. W. Martin 3048, 3067*. Brazil: São Leopoldo, *J. Rick*, 1930 (2 collections), *J. Rick*, 1931; Parecy Novo, *J. Rick*, 1935 (2 collections).

---

right from *GWM 5058*). FIG. 2. *Bourdotia burtii*. A, B. Portions of fertile hyphae, × 875 (A from *J. Rick*, 1930; B from *JR*, 1931). C–E. Basidia, × 875 (C and D from *JR*, 1931; E from *JR*, 1930). F. Gloeocystidium, × 875 (*JR*, 1930). G. Basidiospores, × 875 (*GWM 3048*). FIG. 3. *Bourdotia aspera*. A, B. Basidia, × 900. C–E. Gloeocystidia, × 900. F. Basidiospores, × 900. All from *L. S. Olive T391*. FIG. 4. *Bourdotia pini*. A, B. Portions of fertile hyphae, × 875 (TRT 13090). C, D. Gloeocystidia, × 875 (C from TRT 13090; D from *E. J. H. Corner 666*). E. Basidiospores, × 875 (TRT 13090). All figures drawn with the aid of a camera lucida.

Bodman (**1952**) excluded both *Heterochaete kneiffiopsis* Pat. and *Heterochaete sublivida* Pat. from the genus *Heterochaete*. Examination of the type specimens reveals that *H. sublivida* should be treated as a member of the genus *Exidiopsis*. The type collection of *H. kneiffiopsis* should not, in my opinion, be transferred to the genus *Exidiopsis*, since sterile hyphal pegs are present. The specimens referred to *B. burtii* are *Heterochaete*-like in appearance; however, microscopic examinations have shown that the tubercles are due to a conical accumulation of mineral substance either piercing the hymenium or over which the hymenium is formed. Bresadola mentions in his description of *H. burtii* conical crystalline masses forming the papillae, which is the case in the collections described here. Other characters of *H. burtii* as described by Bresadola are similar to the characters of the collections referred here to this species.

The structure of the fructification and the characters of the fertile hyphae suggest a relationship with the genus *Bourdotia*. This is further indicated by the presence of gloeocystidia arising from the basal hyphal layer; however, these gloeocystidia rarely develop the conspicuous yellow-granular contents characteristic of most species of *Bourdotia*.

B. burtii is somewhat similar to *B. petiolata*, from which it is easily distinguished by the texture and the characters of the gloeocystidia. The obovate basidia of *B. burtii* are somewhat similar to the clavate basidia of *B. petiolata*, but in the former species the longitudinal septa do not divide the basidia into a stalk and fertile portion as in *B. petiolata*.

4. **Bourdotia aspera** (Olive) comb. nov. FIG. 3

Sebacina aspera Olive, Bull. Torrey Bot. Club **85**: 26. 1958.

Effused, indeterminate, when moist appearing as a light gray, pruinose layer; drying to lighter pruinose layer; margins adnate; fructification 20–35 μ in thickness, consisting of several strands of prostrate hyphae from which arise clusters of fertile hyphae and gloeocystidia; gloeocystidia subcylindrical or subfusiform, at first hyaline, becoming yellow-granular, 16.5–33 × 5–9 μ; probasidia formed in clusters at apex of fertile hyphae, at first subglobose, becoming broadly obovate and 2–4-celled by the formation of longitudinal septa, guttulate, 21–27 × 18–22.5 μ; epibasidia not well-defined, up to 17 μ in length, 4–5 μ in diameter at base; basidiospores subglobose, with blunt apiculus, developing a thick spinose or verrucose wall.[5]

[5] This description has been compiled mainly from Olive's original treatment, after examination of the type specimen, and is modified slightly to emphasize certain features and to conform to the format adopted in this paper.

On dead wood. Known only from the type collection.

Type locality: Fautaua Valley, Tahiti.

Illustration: Bull. Torrey Bot. Club **85**: 25, *f. 11.*

Specimen examined: Tahiti: *L. S. Olive T391* (TYPE, in NY).

A unique species well characterized by the large guttulate basidia and the large, subglobose basidiospores which develop conspicuously thick verrucose to spinose walls.

5. **Bourdotia pini** (Jackson & Martin) comb. nov. FIG. 4

Sebacina (*Bourdotia*) *pini* Jackson & Martin, Mycologia **32**: 684. 1940.

Arid-waxy, broadly effused in broken patches, grayish-hyaline to smoke gray; surface smooth to undulate; margins adnate, indeterminate, farinaceous; drying to an ochraceous to grayish-white crust; fructification 15–120 μ thick, consisting of a horizontal hyphal layer of indistinct agglutinate hyphae from which arise the gloeocystidia, fertile hyphae, and, in some cases, the dikaryophyses; 2 or more growth layers sometimes present, each with a horizontal hyphal layer; gloeocystidia at first hyaline, becoming yellow-granular, often collapsing, numerous, various in shape but usually cylindrical to subclavate, 15–45 × 5–12 μ; dikaryophyses short-branched, becoming indistinct, 1–2 μ in diameter, forming an ill-defined layer 10–20 μ above the basidia, arising from the horizontal hyphal layer or from the fertile hyphae; fertile hyphae 1.5–3.5 μ in diameter, forming basidia in acropetal series, the collapsed basidia persisting to form an involucre-like sheath along the fertile hyphae; basidia at first clavate, then obovate to subglobose, and finally suburniform, becoming 2–4-celled, very conspicuously guttulate, 15–27 × (16.5–) 15–22 μ; epibasidia subulate, divergent, up to 20 μ in length, 3–5 μ in diameter at the base; basidiospores cylindrical and laterally depressed to ovate, conspicuously guttulate, usually with 1 large oil droplet, 12–22 × (7–)8–12 μ, germinating by repetition.

On dead wood. Known from Ontario, Oregon, and Brazil.

Type locality: Maple, Ontario.

Illustrations: Mycologia **32**: 685, *f. 2;* Lloydia **4**: 34, *f. 83–87.*

Specimens examined: North America: Ontario, TRT 13090 (TYPE), 21814; Oregon, *A. M. & D. P. Rogers 393, 428.* Brazil: Rio de Janeiro, *E. J. H. Corner 666.*

B. pini is easily recognized by its large basidia and spores, both with large oil droplets; the narrow tortuous fertile hyphae; and the finely branched dikaryophyses.

Although no completely mature basidia were found in the collection from Brazil, there is little doubt of its identity.

6. BOURDOTIA CINEREA (Bres.) Bourd. & Galz., Hymén. Fr. 49. 1928.
Sebacina cinerea Bres., Fung. Trid. **2**: 99. 1892.

This species was described and illustrated in Lloydia **20**: 56, *f. 8*. 1957. Olive (**1958**) has recently reported it from Tahiti, indicating that it is there quite common.

During the early stages of this study I was convinced that the type of *Sebacina rimosa* Jackson & Martin (Martin, **1940**) represented a distinct species; however, after studying additional specimens, especially those from Ontario and Oregon, I have concluded that the type of *S. rimosa* is a thin specimen of *B. cinerea* with unusually globose spores and basidia. Certain specimens, such as *A. M. & D. P. Rogers 551*, are intermediate between the "typical" temperate specimens of *B. cinerea* and the type of *S. rimosa*. Accordingly, I should now consider *S. rimosa* as a later synonym of *B. cinerea*.

Taking into account Olive's description and the additional specimens I have examined, it appears desirable to modify the basidial size to 8–20(–25) × 7–12.5(–13.5) μ and the basidiospore size to 7–12 × 4–10 μ. In addition, several of the collections were 300 μ or more in thickness.

Additional specimens examined: North America: Ontario, TRT 13086 (TYPE of *Sebacina rimosa* Jackson & Martin), 16835; Oregon, *A. M. & D. P. Rogers 551;* California, *K. Wells 312*. Panamá: Barro Colorado Island, *G. W. Martin & A. L. Welden 7898*. Brazil: São Leopoldo, *J. Rick*, 3 collections; Salvador, *J. Rick*, Oct., 1939. Marshall Islands, *D. P. Rogers 1580*. Hawaii: *DPR 1925*.

7. BOURDOTIA CAESIO-CINEREA (Höhn. & Litsch.) Bourd. & Galz. ex Pilát & Lindtner, Glasnik, Bull. Soc. Sci. Skoplje **18**: 175. 1938. FIG. 5

Corticium caesio-cinereum Höhn. & Litsch., K. Akad. Wiss. Wien Sitzungsber. Math.-Naturw. Kl. I **117**: 1116. 1908.

Gloeocystidium caesio-cinereum (Höhn. & Litsch.) Bourd. & Galz., Bull. Soc. Myc. Fr. **28**: 369. 1913.

Bourdotia cinerella Bourd. & Galz., Bull. Soc. Myc. Fr. **36**: 71. 1920.

Sebacina cinerella (Bourd. & Galz.) Killerm., in Engl. & Pr., Nat. Pflanzenfam. 2 ed. **6**: 115. 1928.

Sebacina caesio-cinerea (Höhn. & Litsch.) Rogers, Univ. Iowa Stud. Nat. Hist. **17**: 37. 1935.

Arid-waxy, effused, indeterminate, whitish to pale gray; surface pruinose or porous-reticulate to nearly continuous, in some cases forming

scattered fertile columns; margins completely adnate; drying to a light-gray pruinose layer; fructification 25–75(–90) μ in thickness, consisting of a basal layer of agglutinate hyphae arranged parallel with the substrate, usually thin and obscure, giving rise directly to the fertile hyphae and gloeocystidia; sometimes two or more growth layers present, each consisting of a horizontal hyphal layer and the ascending elements; gloeocystidia cylindrical, subfusiform, or subclavate, at first hyaline, soon becoming yellow-granular and collapsing, 10–40 × 4–8.5 μ, sometimes with expanded globose heads up to 15 μ in diameter; fertile hyphae erect, tortuous, 1.5–3.5 μ in diameter, forming basidia in a close acropetal series, in older specimens ensheathed by the collapsed basidia and bearing 2–3 probasidia or mature basidia at the apex; probasidia at first obovate, becoming ovate to suburniform and 2–4-celled, 10–17(–18.5) × 7–12.5 (–13.5) μ; epibasidia not apparent, sterigmata subulate, 5–11 μ in length and 1.5–3 μ in diameter at the base, often divergent; basidiospores subglobose with prominent apiculi, 1–2 μ in length, symmetrically attached, often guttulate, (5–)6–11 μ in diameter, sometimes minutely spinose.

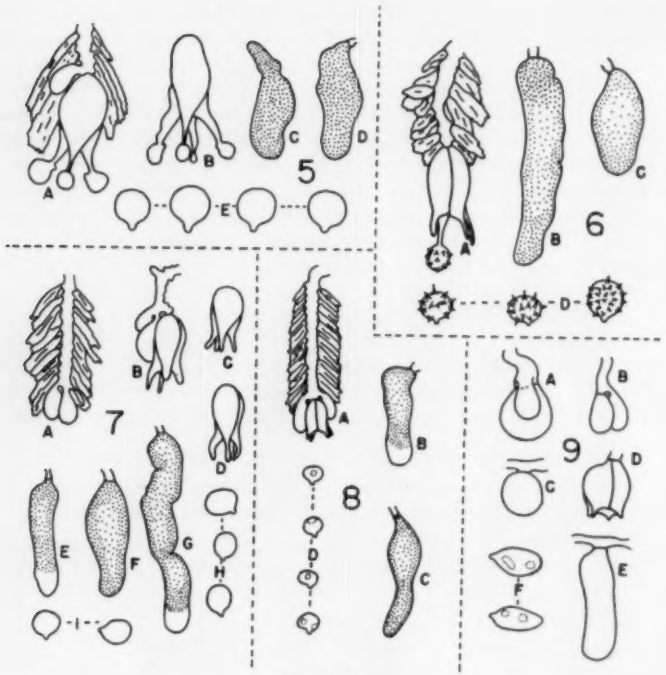
On very rotten wood, usually decorticated. Known from Ontario, Quebec, British Columbia; eastern United States, south to Florida and west to Iowa and Missouri; Oregon; Europe; and North Africa.

Type locality: Austria.

Illustrations: Bourd. & Galz., Hymén. Fr. 49, *f. 27;* Univ. Iowa Stud. Nat. Hist. **15**: *pl. 1, f. 10–12;* Lloydia **4**: 38, *f. 106–108;* Bull. Soc. Myc. Fr. **70**: 123, *f. 1, d.*

Specimens examined: Canada: Ontario, TRT 8498, 11097, 16848; Quebec, TRT 11131; British Columbia, *R. J. Bandoni BC-133* (in UBC). United States: Massachusetts, *D. H. Linder,* 20 April, 1936, *D. P. Rogers 2241;* Rhode Island, *A. M. & D. P. Rogers 2248;* Florida, *R. Singer F-1038;* Iowa, *G. W. Martin 6300;* Oregon, *AM & DPR 257, 1052, 2249, 2250, 2251, 2252, 2253, 2254, 2255, 2256, 2264.* Sweden: *S. Lundell,* 28 Oct., 1929, *1190, S. Lundell & G. Haglund 5854, J. Eriksson 6484.*

This species is characterized by the subglobose basidiospores with prominent apiculus, the dense fructification with numerous gloeocystidia, and the usually ovate basidia with subulate sterigmata. The spore walls are often minutely spinose. *B. caesio-cinerea* is apparently closely related to *B. eyrei,* from which it may be distinguished by the larger subglobose basidiospores and the symmetrically attached apiculi, which usually attain a length of 1–2 μ, the usually shorter and broader gloeocystidia, and the larger basidia. The basidiospores of *B. eyrei* may have somewhat prominent apiculi; however, they are always obliquely attached.



FIGS. 5–9.

FIG. 5. *Bourdotia caesio-cinerea.* A. Portion of fertile hypha, × 875 (*D. H. Linder,* 20 April, 1936). B. Basidium, × 875 (*S. Lundell & G. Haglund 5834*). C, D. Gloeocystidia, × 875 (*G. W. Martin 6300*). E. Basidiospores, × 875 (two left spores from *J. Eriksson 6484;* two right spores from *S. Lundell,* 28 Oct., 1929). FIG. 6. *Bourdotia spinosa.* A. Portion of fertile hypha, × 900. B, C. Gloeocystidia, × 900. D. Basidiospores, × 900. All from *L. S. Olive T398.* FIG. 7. *Bourdotia cyrei.* A, B. Portions of fertile hyphae, × 875 (A from *G. W. Martin 1491;* B from *A. H. Smith 13402*). C, D. Basidia, × 275 (C from *GWM 1483;* D from *AHS 13402*). E–G. Gloeocystidia, × 875 (E and F from *AHS 13402;* G from *S. Lundell & G. Haglund 5682*). H, I. Basidiospores, × 875 (H from *GWM 1491;* I from *GWM,* 24 April, 1940). FIG. 8. *Bourdotia grandinioides.* A. Portion of fertile hypha, × 875 (Ell. & Everh., Fung. Col. 1017). B, C. Gloeocystidia, × 875 (B from *D. P. Rogers 436;* C from E. & E., Fung. Col. 1017). D. Basidiospores, × 875 (from top to bottom, from E. & E. Fung. Col. 1017, from *D. H. Linder 691,* remainder from *DPR 436*). FIG. 9. *Bourdotia obscura.* A–D. Basidia in various stages of development, × 875 (A and B from *G. W. Martin & A. L. Welden 7638;* C and D from *GWM 2873*). E. Gloeocystidium, × 875 (*GWM & ALW 7638*). F. Basidiospores, × 875 (*GWM & ALW 7638*). All figures drawn with the aid of a camera lucida.

8. **Bourdotia spinosa** (Olive) comb. nov. FIG. 6

Sebacina spinosa Olive, Bull. Torrey Bot. Club **85**: 27. 1958.

Effused, indeterminate, when moist appearing as a thin farinaceous layer; drying to a light gray or tan farinaceous layer; margins adnate; fructification 30–50 μ in thickness, consisting of a few strands of prostrate hyphae, from which arise the isolated strands of fertile hyphae and gloeocystidia; gloeocystidia at first hyaline, becoming yellow-granular and flexuous at maturity, subcylindrical to subfusiform, 18–50 × 5–11 μ, fertile hyphae forming basidia in acropetal series, the collapsed basidia persisting as an involucre-like sheath along the axis of the fertile hyphae; basidia becoming subglobose, ovate, or suburniform and cruciately septate, 9–10 × 7.5–9 μ; epibasidia not differentiated, sterigmata up to 10 μ in length, 1–2 μ in diameter at base, often adaxially recurved; basidiospores subglobose, sometimes with short axis through the prominent apiculus, developing a thick spinose wall, (4.5–)5–8 μ in diameter.[5]

On rotten wood. Known only from the type collection.

Type locality: Fautaua Valley, Tahiti.

Illustration: Bull. Torrey Bot. Club **85**: 25, *f. 12.*

Specimen examined: Tahiti: *L. S. Olive T398* (TYPE, in NY).

A species similar to *B. caesio-cinerea* in many respects, but distinguished by the development of the very thick spinose walls on the basidiospores.

9. BOURDOTIA EYREI (Wakef.) Bourd. & Galz., Hymén. Fr. 50. 1928. FIG. 7

Sebacina eyrei Wakef., Brit. Myc. Soc. Trans. **5**: 126. 1915.

Gloeocystidium croceo-tingens Bres. (attributed to Wakef.), Ann. Myc. **18**: 48. 1920.

Sebacina deminuta Bourd., Ass. Fr. Av. Sc. **45**: 575. 1922.

Gloeocystidium eyrei (Wakef.) Sacc., Trev., & Trott., Syll. Fung. **23**: 518. 1925.

Corticium involucrum Burt, Ann. Missouri Bot. Gard. **13**: 271. 1926.

Bourdotia deminuta (Bourd.) Bourd. & Galz., Hymén. Fr. 50. 1928.

Bourdotia rimulenta Bourd. & Galz., Hymén. Fr. 51. 1928.

Bourdotia mucosa Bourd. & Galz., Hymén. Fr. 51. 1928.

Basidiodendron luteo-griseum Rick, Brotéria **25**: 74. 1938.

Waxy, effused, at first light gray, becoming light ochraceous to reddish brown; drying to a pruinose, porous-reticulate or continuous layer, becoming cinereous, pinkish-gray or light reddish-brown; margins farinaceous to fimbriate, adnate and indeterminate, usually somewhat lighter;

fructification 17–125(–180)μ in thickness, consisting of a thin basal layer of agglutinate hyphae parallel to the substrate, from which arise the gloeocystidia and fertile hyphae, in some specimens up to 5 growth layers present, each consisting of a horizontal hyphal layer which gives rise to gloeocystidia and fertile hyphae; gloeocystidia cylindrical to subclavate or subfusiform, at first hyaline, becoming yellow-granular in an acropetal direction, collapsing at maturity, often with hyaline apex, sometimes extending through 2 or more growth layers, 10–65 × (3.5–)4–7.5(–8.5)μ, rarely with globose apex up to 12μ in diameter; fertile hyphae forming basidia at the surface of the fructification in a close acropetal series, tortuous, 1.5–3μ in diameter, in older specimens surrounded by a dense involucre-like sheath of collapsed basidia and bearing at the apex a cluster of probasidia or mature basidia; probasidia arising as obovate to subglobose structures, becoming ovate to urniform and cruciately septate, sometimes guttulate, (7–)8.5–12.5 × (5–)6–8.5μ; epibasidia poorly differentiated, subulate, adaxially recurved or divergent, up to 9μ in length, 1.5–2μ in diameter at the base; basidiospores subglobose, broadly elliptical, broadly obovate to elongate, usually guttulate, minutely apiculate or with blunt apiculus obliquely attached, 4.5–7 × (3.5–)4–6μ, germinating by repetition.

On decaying wood, usually decorticated. Known from Ontario; Quebec; eastern United States, west to Washington, Oregon, and California; Panamá; Colombia; Brazil; Hawaii; Tahíti; Europe, and North Africa. Probably cosmopolitan.

Type locality: England.

Illustrations: Bourd. & Galz., Hymén. Fr. 50, *f. 28;* Univ. Iowa Stud. Nat. Hist. **15**: 25, *f. 7–9, f. 13–16;* **18**: 77, *f. 12;* **19**: *pl. 2, f. 12;* Mycologia **27**: 506, *f. 1–55;* Lloydia **4**: 38, *f. 95–99, f. 100–105;* Bull. Soc. Myc. Fr. **70**: 123, *f. 1, d.*

Specimens examined: Canada: Ontario, *D. H. Linder,* 2 May, 1936, TRT 16846, 16849; Quebec, *I. Mounce,* 1 Oct., 1935. United States: Massachusetts, *G. D. Darker 4791, A. M. & D. P. Rogers 547;* New York, *AM & DPR 561;* New Jersey, *G. W. Martin,* 10 Sept., 1932; Ohio, *D. P. Rogers,* Sept., 1933; Iowa, *GWM 1383, 1482, 1491, AM & DPR 317,* and 2 other collections; Oregon, *AM & DPR 561, DPR 564;* Washington, *A. H. Smith 13402;* California, *K. Wells 239, 286, 298, 303, 308, 398.* Panamá: Barro Colorado Island, *G. W. Martin & A. L. Welden 7059, 7589, 7680, 8409, 8415, 8525, 8571;* Valley of Rio Chagres, *GWM 3062;* Prov. Chiriquí, *GWM 2372, 3009, 4458;* Canal Zone, *GWM 6023.* Colombia: Sierra Nevada de Santa Marta, *GWM 3523, 3550.* Brazil: São Leopoldo, *J. Rick 54, J. Rick,* Sept. 1939; Parecy Novo, *J. Rick,* 1935 (authentic specimen of *Basidiodendron*

luteo-griseum Rick, in FH); Sta. Maria, *J. Rick*, 1936; Rio de Janeiro, *E. J. H. Corner 665, 669.* Hawaii: Oahu, *E. A. Bessey 586, DPR 1189, 1190, 1192, 1193, 1261, 1843, 1845, 1846, 1866.* Sweden: *S. Lundell & G. Haglund 5682, S. Lundell 2014.* England: *W. Eyre,* 7 May, 1914 and *C. A. Cooper,* Oct., 1914 (SYNTYPES of *Sebacina eyrei* Wakef., in K). France: *Galzin 10159, 13279* (SYNTYPES of *Bourdotia rimulenta* Bourd. & Galz., in PC), *Galzin 5428* (HOLOTYPE of *Bourdotia mucosa* Bourd. & Galz., in PC), *Galzin 14773* (authentic specimen of *Sebacina deminuta* Bourd., in PC). Austria: Tirol, *V. Litschauer, 35, 36.*

This species is somewhat intermediate between *B. caesio-cinerea* and *B. grandinioides* in basidia and basidiospore size and structure. The spores of *B. eyrei* never have the shortest axis through the apiculus as in *B. grandinioides* nor do they possess the blunt apiculus which is symmetrically attached as in *B. caesio-cinerea.* In addition, there are rather consistent differences in size of the mature basidia among the three species.

McGuire (**1941**) points out that the distinction between *B. eyrei* and *B. deminuta* may be an arbitrary one; however, since he was unable to examine the type collections of the two species, he did not combine them. Olive (**1958**) noted that the difference in spore shape, previously used to separate the two taxa, was not constant and accordingly combined them. Although the two specimens upon which Wakefield based *Sebacina eyrei* have predominantly subglobose spores and a specimen which has been examined by H. Bourdot and determined by him as *Sebacina deminuta* has mainly oblong to broadly elliptical spores, there are in these three collection examples of basidiospores which vary from subglobose to broadly elliptical to oblong. Several other collections possess spores which vary from broadly elliptical or broadly obovate to subglobose; therefore, I have followed Olive's treatment of the two taxa, especially in view of the fact that there is no discernible morphological difference in the structure of the fructifications or in the structure and size of the basidia.

The two specimens of *B. rimulenta* upon which the original description was based and the holotype of *B. mucosa* appear to be only thin specimens of *B. eyrei,* differing somewhat in macroscopic appearance, but microscopically quite similar to the syntypes of *Sebacina eyrei.*

A specimen in the Farlow Herbarium labelled by Rick as *Basidiodendron luteo-griseum* Rick agrees in all respects with the characters of the other specimens assigned to this species; therefore, *B. luteo-griseum* is considered here as an additional synonym of *B. eyrei.*

10. BOURDOTIA GRANDINIOIDES Bourd. & Galz., Hymén. Fr. 51. 1928. FIG. 8

Sebacina grandinioides (Bourd. & Galz.) Rogers, Univ. Iowa Stud. Nat. Hist. **17**: 40. 1935.

Arid-waxy, effused; surface pruinose to porous-reticulate, with scattered obtuse columns arising from the substrate; margins adnate, indeterminate and usually pruinose; drying to a whitish pruinose, light gray, or light buff layer; fructification 20–45 μ in thickness and often with fertile hyphal columns arising up to 200 μ from the substrate, in thinner specimens consisting of a few strands of prostrate hyphae, from which arise directly the fertile hyphae and gloeocystidia, at the margins the basidia sometimes borne directly on the prostrate hyphae, in thicker specimens with vertical strands of hyphae bearing radially gloeocystidia and fertile hyphae forming the macroscopically visible columns; gloeocystidia at first hyaline, soon becoming yellow-granular and flexuous, cylindrical, subfusiform, or ventricose, usually quite abundant, 12–50 × 2–7(–8) μ; fertile hyphae forming basidia in a close acropetal series, flexuous, 1.5–2.5 μ in diameter, in thicker specimens collapsed basidia forming an involucre-like sheath along the fertile hyphae, which bear clusters of 2–3 basidia at the apex; probasidia arising as obovate to subglobose structures, becoming ovate to suburniform and 2–4-celled, 6.5–8(–9) × 5–7 μ; epibasidia not differentiated, sterigmata divergent or incurved, subulate, up to 5 μ in length, and approximately 1 μ in diameter at the base; basidiospores elliptical to subglobose, usually with the shortest axis through the minute apiculus, often guttulate, 3–5 × 3–4 μ.

On decaying wood. Known from Ontario, New Jersey, Oregon, British Guiana, and France.

Type locality: France.

Illustrations: Bourd. & Galz., Hymén. Fr. 51, *f. 29;* Univ. Iowa Stud. Nat. Hist. **17**: 35, *pl. 3, f. 18;* Lloydia **4**: 38, *f. 109–112.*

Specimens examined: Ontario: TRT 16844, 16845; New Jersey: Ellis & Everh. Fung. Col. 1017 (as *Hydnum farinaceum* var. *luxurians* Cke.); Oregon: *D. P. Rogers 436, AM & DPR 2267;* British Guiana: *D. H. Linder 691.*

Six additional specimens have been assigned to this species since McGuire's (**1941**) treatment. The collection examined by McGuire (Ellis & Everh. Fung. Col. 1017) is considerably thicker than the additional collections, with well-developed fertile columns present. In one Oregon collection (*DPR 436*) there is definite evidence of fertile columns, since the basal hyphal layer is thickened in certain areas; however, they do not attain the size of the columns in the New Jersey collection.

The collection from British Guiana is quite thin and appears as a white porous-reticulate layer in the older portions, with the margins thinning out to a white pruinose area. Near the margins, the fertile hyphae have no involucre-like sheath of collapsed basidia, and frequently the basidia are borne directly on the prostrate hyphae without any visible fertile hyphae. In these regions, the gloeocystidia are hyaline or, at most, faintly yellow. Thus, the thinner specimens of *B. grandinioides* are quite similar to the known collections of *Sebacina obscura*—in both micro- and macroscopic aspects.

11. **Bourdotia obscura** (Martin) comb. nov. FIG. 9

Sebacina obscura Martin, Lloydia **7**: 70. 1944.

Widely effused, indeterminate, very thin, when moist appearing as a white, arachnoid growth over a grayish, gelatinous film; when dry appearing as a white, arachnoid to pruinose layer over a gray to light brown film; margins adnate, similar to mature portions; fructification 10–30 μ thick, consisting of several prostrate strands of hyphae 1.5–3 μ in diameter, without observable clamp-connections, from which arise the gloeocystidia and fertile hyphae; gloeocystidia cylindrical, obtuse, hyaline, widely scattered, 15–30 × 5–7 μ; probasidia 6–10 × 5.5–8.5 μ, borne either on repent hyphae or on short, upright, fertile hyphal segments 2.5–3 μ in diameter, proliferating from the base of the basidia without clamp-connections, at first obovate, becoming globose and cruciately septate, then appearing truncate at the apex; epibasidia not differentiated, each basidial segment producing directly a short, slender sterigma upon which a basidiospore is borne; basidiospores elliptical-fusoid, somewhat curved, often guttulate, 9–10 ×23.5–4.5 μ.

Reported only on dead attached leaf stalks of the date palm. Known only from Summit and Barro Colorado Island in Panamá.

Type locality: Summit, Panamá Canal Zone.

Illustration: Lloydia **7**: 69, *f. 5;* Lloydia **20**: 60, *f. 11.*

Specimens examined: Panamá: Canal Zone, Summit, *G. W. Martin 2873* (TYPE); Barro Colorado Island, *G. W. Martin & A. L. Welden 7638.*

Martin (**1944**) in his discussion of this species suggested that it may be related to the arid species of *Bourdotia,* especially *B. grandinioides* and *B. eyrei;* however, since the type collection is quite thin with most of the basidia borne on the prostrate hyphae and with gloeocystidia only very rarely formed, the species was not placed in the section *Bourdotia.* The single additional collection (*Martin & Welden 7638*) reported by myself (**1957**) was somewhat thicker, with scattered hyaline gloeocys-

tidia and with basidia often borne on fertile hyphae arising from the prostrate hyphae. However, because of the lack of an involucre-like sheath and yellow-granular gloeocystidia, I did not feel justified in transferring *S. obscura* to *Bourdotia*. Additional study has shown that the early stages of *B. grandinioides* are essentially similar to the known collections of *B. obscura;* i.e., in *B. grandinioides* the basidia are initially borne on the prostrate hyphae and the gloeocystidia are scattered and frequently hyaline. The basidia in both species are quite similar in that both lack epibasidia, appear somewhat truncate at the apex, and are formed in dense clusters by proliferations of the fertile hyphae immediately below the older basidia. Further, there is no fundamental difference in the structure of the younger fructifications, since both species possess a thin layer of prostrate hyphae from which arise the fertile hyphae or basidia and the gloeocystidia. Thus, it would appear to be logical to consider the two collections of *B. obscura* as representing a very primitive or very much reduced member of the genus *Bourdotia*. It is also conceivable that this species is related to *Heterochaetella dubia* (Bourd. & Galz.) Bourd. & Galz. and *Sebacina minima* Olive as Olive (**1958**) has suggested; however, the exact basidial structure of *H. dubia* is not known, since these structures collapse upon drying and cannot be revived, a character which I have also noted in the type specimen of *S. minima*.

SPECIES DUBIA

1. SEBACINA CARNEOLA Bresadola, Stud. Trent. II **7**: 64 (= Sel. Myc. II. 14). 1926. (From Bourd. & Galz., Hymén. Fr. 46. 1928.)

The author indicates that this species is a relative of *Bourdotia cinerea;* however, he does not mention the presence of gloeocystidia.

SPECIES EXCLUDENDAE

1. SEBACINA MUCEDINEA Pat., Bull. Herb. Boiss. **3**: 60. 1895.

Olive (**1958**) includes this species in the section *Bourdotia* (sensu McGuire) because of the presence of inflated hymenial elements. In my opinion these elements are homologous with such structures found in *Exidiopsis grisea* (Pers.) Bourd. & Maire, *Exidiopsis fuliginea* Rick, and others, not with the gloeocystidia of the genus *Bourdotia,* which develop a yellow-granular *content* and which arise from the basal hyphal layer. In several collections from South America, I (**1957**) have found that specimens referred to *Exidiopsis mucedinea* (Pat.) Wells have a

well-defined ascending hyphal layer. In addition, basidial development is typical of the genus *Exidiopsis* rather than *Bourdotia*.

2. SEBACINA PUNAAUIAE Olive, Bull. Torrey Bot. Club **85**: 26. 1958.

This species has also been included in the section *Bourdotia* by Olive. The fertile hyphae in the type specimen (*L. S. Olive T427*, in NY) are not typical of the genus *Bourdotia;* i.e., basidial development is by lateral proliferation, resulting in several near-mature basidia at approximately the same level rather than an acropetal series of basidia, as in the genus *Bourdotia*. Basidial development in *Bourdotia* is, of course, by lateral proliferation beneath the youngest basidium, but the older basidia are forced aside, resulting in fertile hyphae which have an essentially straight axis (see Rogers, **1935**, pp. 42–43). I am unable to see clearly the remaining features in the only specimen of *S. punaauiae*, but there appear to be indications of hyphal pegs. Thus, I tentatively suggest that it belongs in the genus *Heterochaete*.

3. SEBACINA UMBRINA Rogers, Univ. Iowa Stud. Nat. Hist. **17**: 39. 1935.

This species was placed by Rogers in the subgenus *Bourdotia* and by McGuire (**1941**) in the section *Bourdotia*. The type specimen (*D. P. Rogers 278*) has an ascending hyphal layer, the fertile hyphae are not typical of the other species included in *Bourdotia*, and the inflated hymenial elements do not become yellow-granular and arise either from the fertile hyphae or from the hyphae bearing the branching dikaryophyses. The specimen should, I think, be referred to *Exidiopsis*—very probably to the species most frequently termed *Sebacina grisea* (Pers.) Bres. [= *Exidiopsis grisea* (Bres.) Bourd. & Maire].

SUMMARY

1. The genus *Bourdotia* is treated as a distinct genus composed of eleven known species, with three species, which have been included in the group by other authors, excluded, and one species treated as doubtful, since authentic material was not examined.

2. *Bourdotia* is characterized by a resupinate habit; horizontal and vertical indeterminate growth; possession of tremellaceous basidia formed in close acropetal series by the proliferation of the fertile hyphae beneath the youngest basidium, forming an essentially straight strand of fertile hyphae which is surrounded by the collapsed basidia; and a structure

composed of a prostrate basal hyphal layer giving rise to gloeocystidia with a yellow-granular content, fertile hyphae, and, in some cases, dikaryophyses.

3. Most of the specimens examined were collected in North and South America; however, a number of collections were available from Europe and certain Pacific Islands. Collections of most species are yet too few to accurately determine the geographic range; nevertheless, it appears that *B. petiolata* is restricted to tropical and subtropical regions, whereas *B. eyrei* and *B. cinerea* are probably cosmopolitan in distribution.

DEPARTMENT OF BOTANY
STATE UNIVERSITY OF IOWA
IOWA CITY, IOWA

LITERATURE CITED

1. **Bodman, M. C.** 1952. A taxonomic study of the genus *Heterochaete*. Lloydia **15**: 193–233.
2. **Bourdot, H., & A. Galzin.** 1928. Hyménomycètes de France, pp. 48–51. Lons-le-Saunier.
3. **Bresadola, J.** 1908. Fungi aliquot gallici novi vel minus cogniti. Ann. Myc. **6**: 46.
4. **Burt, E. A.** 1915 The Thelephoraceae of North America. V. *Tremellodendron, Eichleriella,* and *Sebacina*. Ann. Missouri Bot. Gard. **2**: 749–766.
5. ——. 1926. The Thelephoraceae of North America. XV. Conclusion, with supplement and general index. Ann. Missouri Bot. Gard. **13**: 334–340.
6. **Ervin, M. D.** 1957. The genus *Sebacina*. Mycologia **49**: 118–123.
7. **Gäumann, E. A., & C. W. Dodge.** 1928. Comparative morphology of fungi, pp. 520–528. New York.
8. **Killermann, S.** 1928. Tremellineae. In Engler und Prantl, Die natürlichen Pflanzenfamilien. 2 ed. **6**: 111–119.
9. **Lanjouw, J., & F. A. Stafleu.** 1956. Regnum vegetabile 6. Index herbariorum 1. The herbaria of the world. 3 ed.
10. **Malençon, G.** 1954. Prodrome d'une flora mycologique du Moyen-Atlas. II. Bull. Soc. Myc. Fr. **70**: 118–126.
11. **Martin, G. W.** 1944. New or noteworthy tropical fungi. III. Lloydia **7**: 67–80.
12. ——. 1940. Some Heterobasidiomycetes from Eastern Canada. Mycologia **32**: 683–695.
13. ——. 1952. Revision of the North Central Tremellales. Univ. Iowa Stud. Nat. Hist. **19**: 1–122.
14. **McGuire, J. M.** 1941. The species of *Sebacina* (Tremellales) of temperate North America. Lloydia **4**: 1–43.
15. **Neuhoff, W.** 1936. Die Gallertpilze Schwedens. Arkiv för Bot. **28A**[1]: 1–57.
16. **Olive, L. S.** 1958. The lower Basidiomycetes of Tahiti. Bull. Torrey Bot. Club **85**: 5–27; 89–110.
17. **Rea, C.** 1922. British Basidiomycetae, pp. 737–738. Cambridge.

18. **Rogers, D. P.** 1933. Some noteworthy fungi from Iowa. Univ. Iowa Stud. Nat. Hist. **15**: 9–29.
19. ——. 1935. Notes on the lower Basidiomycetes. Univ. Iowa Stud. Nat. Hist. **17**: 3–43.
20. **Torrend, C.** 1913. Les Basidiomycètes des environs de Lisbonne et de la région de S. Fiel (Beira Baixa). Brotéria sér. bot. **11**: 86–89.
21. **Wells, K.** 1957. Studies of some Tremellaceae. Lloydia **20**: 43–65.
22. ——. 1958. Studies of some Tremellaceae. II. The genus *Ductifera*. Mycologia **50**: 407–416.
23. **Wittlake, E. B.** 1938. Hymenial organization of *Sebacina calcea*. Univ. Iowa Stud. Nat. Hist. **17**: 351–361.

NOTES ON BOLETES. XI.

WALTER H. SNELL, ROLF SINGER AND ESTHER A. DICK

(WITH 1 FIGURE)

I. COLLECTIONS BY SINGER IN MICHIGAN IN 1953

1. STROBILOMYCETACEAE

Strobilomyces floccopus (Vahl in Fl. Dan. ex Fr.) Karst., *Porphyrellus gracilis* (Pk.) Sing., *P. pseudoscaber* (Secr.) Sing., and *Boletellus Russellii* (Frost) Gilb.

2. BOLETACEAE

A. SUBFAMILY GYRODONTOIDEAE

Gyroporus cyanescens (Bull. ex Fr.) Quél., *G. castaneus* (Bull. ex Fr.) Quél., and *Gyrodon merulioides* (Schw.) Sing.

B. SUBFAMILY SUILLOIDEAE

Boletinus pictus (Pk.) Pk., *Suillus acidus* (Pk.) Sing., *S. americanus* (Pk.) Snell, *S. cothurnatus* Sing., *S. granulatus* (L. ex Fr.) Kuntze subsp. *Snellii* Sing., *S. luteus* (L. ex Fr.) S. F. Gray, *S. placidus* (Bon.) Sing., *S. piperatus* (Bull. ex Fr.) Kuntze, *S. punctipes* (Pk.) Sing., *S. subaureus* (Pk.) Snell, and *S. subluteus* (Pk.) Snell.

C. SUBFAMILY XEROCOMOIDEAE

Phylloporus rhodoxanthus (Schw.) Bres. subsp. *albomycelinus* Sing., *Xerocomus badius* (Fr.) Kühner ex Gilb., *X. chrysenteron* (Bull. ex Fr.) Quél., *X. illudens* (Pk.) Sing., and *X. subtomentosus* (L. ex Fr.) Quél.

D. SUBFAMILY BOLETOIDEAE

Boletus edulis Bull. ex Fr. subsp. *clavipes* Pk., *B. griseus* Frost subsp. *griseus, B. luridus* Schaeff. ex Fr., *B. pallidus* Frost, *B. rubellus* Krombh. subsp. *fraternus* (Pk.) Sing., *B. speciosus* Frost, *B. subvelutipes* Pk., *B. vermiculosus* Pk., *Tylopilus felleus* (Bull. ex Fr.) Karst.,

Xanthoconium affine (Pk.) Sing., *Leccinum aurantiacum* (Bull. ex) S. F. Gray, *L. chromapes* (Frost) Sing., *L. oxydabile* (Sing.) Sing., *L. scabrum* (Bull. ex Fr.) S. F. Gray subsp. *scabrum* and subsp. *niveum* (Fr.) Sing., *L. subglabripes* (Pk.) Sing., *L. testaceoscabrum* (Secr.) Sing.

Most of the species enumerated above are either sufficiently well known to justify omission of their descriptions, or were previously described in The Boletineae of Florida, I–III.[1] A few species, however, should be redescribed.

II. REDESCRIPTIONS

SUILLUS ACIDUS FIG. 1A

Pileus basically whitish to pallid (Ivory to Sheepskin[2]), marked with pigmented dots, spots, streaks or areas, with the result that the general color is either whitish or more or less chamois-colored (Chamois, Buttercup or Syrup), glabrous, viscid, pulvinate, 4–7 cm broad. *Hymenophore* tubulose, with tubes and pores more or less as in *S. granulatus*. *Stipe* white to yellowish-white, sometimes stained gray, covered both above and below the annulus with glandules which appear as dots, specks or streaks initially Colonial Buff, later sordid gray and eventually black; attenuated upward, eventually equal, 55–75 × 9–12 mm; annulus when present apical and narrow, well-developed, belt-like or fragmentary, simple, not sheathing, often whitish when fresh, light yellow to olive-yellow in age, or sometimes completely absent, in which case the margin of the pileus will always be appendiculate with traces of the veil; mycelial tomentum whitish to Buttercup or Banana. *Context* of the pileus at first whitish-yellow, the upper portion of the stipe lemon-yellow and the base orange but soon, as in *S. subluteus*, everywhere pale orange except for deep orange marbling (Doubloon to Punjab) in the stipe, unchanging in dry weather; odor pleasant, as in *S. granulatus*, but weak; taste characteristically strongly acid and extremely unpleasant, as sour as H_2SO_4.

Microscopic characters much like those of *S. subluteus*.

KOH on pileus producing red dots and deep bluish-gray discoloration; on young context at first negative, later mostly somewhat violet. NH_4OH on young flesh negative to somewhat violet; on pores negative.

[1] Rolf Singer. The Boletineae of Florida with notes on extralimital species. I. The Strobilomycetaceae. Farlowia **2**: 97–141. 1945. II. The Boletaceae (except Boletoideae). Farlowia **2**: 223–303. 1945. III. The Boletoideae of Florida. Am. Mid. Nat. **37**: 1–126. 1947.

[2] Color terms with a capital letter are those of Maerz and Paul, Dictionary of Color, 1950, unless designated by R, for those of Ridgway's Color Standards and Color Nomenclature, 1912.

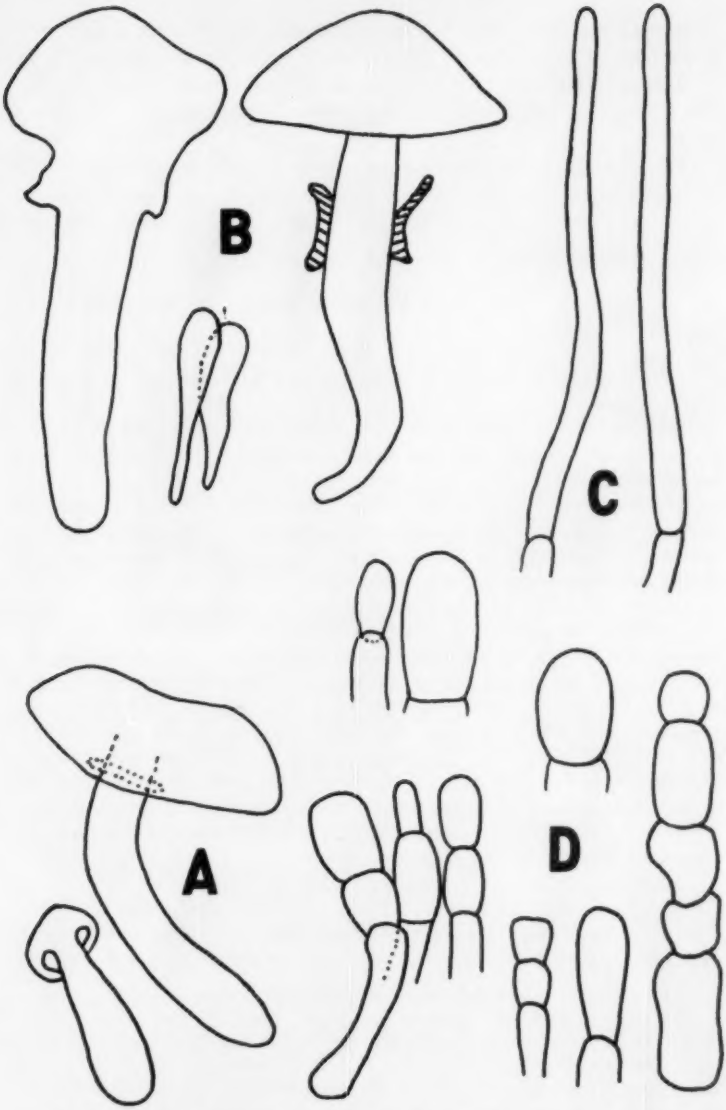


FIG. 1. A. *Suillus acidus.* B. *Suillus cothurnatus,* with cystidia. Elements of the cuticle of the pileus, × 800—C, of *Leccinum scabrum* subsp. *scabrum,* and D, of *L. oxydabile.*

On the ground under *Pinus resinosa* exclusively. Douglas Lake, Cheboygan County, Mich.

SUILLUS SUBAUREUS and S. AMERICANUS

These species have been described before, but inasmuch as they are very close, we present a comparison of their characters rather than formal redescriptions.

Suillus subaureus	*Suillus americanus*
Pileus a mixture of Chinese Yellow, Brass and Yellow Ochre, or with a Narcissus ground color, when dry usually with a Spruce Yellow or Madura cast and then completely covered with grayish to brownish-gray, well-attached, rough veil fragments which are more Popcorn color or paler at the margin, the young caps more olive-gray (13F5) and with often persistent, tiny, cinnabarinous (Brick Red) specks; subviscid or viscid, somewhat scaly in the center, with conglutinated fibrils at the margin, but hardly really uneven, convex.	Pileus Aureoline, Sunflower, Light Chrome or 10L3, the young margin often appendiculate with crenate velar fragments, otherwise occasionally with scattered, yellow, fibrillose remains of the veil which are later brown (Cocoa), also with occasional bright red spots when young, fresh and dry; very viscid to glutinous, soon quite glabrous, even, convex, often umbonate.
Hymenophore tubulose, 3.5–4.5 mm thick in a pileus 35 mm broad, the pores Apricot Yellow to Forsythia, then Jasmine or even duller, also yellow within, not gyrose when young, small, later rather wide (about 1 mm).	Hymenophore tubulose, 4.5 mm thick in a pileus 45 mm broad, the pores Primuline Yellow, staining brownish-vinaceous-pink on bruising and within also yellow and staining, 9–10 pores a cm radially, 12–13 a cm tangentially halfway to margin.
Spore print 13H9 to Raw Sienna, near Tiffin in a very thick layer.	Spore print similar in color.
Stipe Lemon Yellow with some Light Chrome, adorned with erect to wart-like glandules which may be dots or short ridges, not sticky, concolorous or somewhat more sordid, eventually directly chang-	Stipe yellow, dotted with glandules that are extremely flat and streak-like, and not suggesting a scabrosity, yellow then cinnamon, then brownish-red and finally blackish-brown; cylindric and rela-

ing to black; *not* relatively thin; basal tomentum white, somewhat yellow near the very base; annulus none.

Context fleshy in the pileus, rather hard-fibrous in the stipe, light yellow, deep yellow in the base of the stipe, staining Sayal Brown at the apex of the stipe when broken; odor agreeable like *S. placidus;* taste slightly acidulous but mild and pleasant.

Spores hyaline, 7–10 × 3–4 μ, mostly 7.5–8.5 × 3 μ.

tively thin as in *S. subluteus,* or thinner; basal tomentum white, extending for 7–11 mm, at very base often pinkish; annulus none.

Context soft-fleshy in the pileus, often rather hard-fibrous in the stipe, lemon-yellow throughout but staining vinaceous-pinkish in the pileus and upper portion of the stipe; odor and taste as in *S. subaureus.*

Spores pale yellow, 8–11 × 3.5–4.5 μ, mostly 9 × 4 μ.

Other microscopic characters nearly identical in both species as far as known.

KOH on pileus black (including red spots); on pores sordid, then chestnut-black; on context deep gray.

NH_4OH slowly bluish-green to blue on context (Ripple Green to Email), from the vapors around the drop rapidly orange and then salmon.

H_2SO_4 on surface of pileus and stipe deep wine-red (Chianti); on context almost no reaction.

Habitat: under *Populus tremuloides* with *or rarely without* conifers in the immediate neighborhood (e.g. *Tsuga canadensis, Pinus* spp.), also in coniferous woods or mixed woods under pines.

KOH on pileus black (including red spots).

NH_4OH Evergreen on the flesh, NH_3 (around drop) bright orange-rufous (Maya to Talavera).

H_2SO_4 on surface of pileus carmine-red, then cadmium yellow.

Under *Pinus* exclusively, especially under *P. strobus.*

SUILLUS COTHURNATUS FIG. 1B

Pileus yellow (Gold Leaf) or pallid with fuliginous gluten, the latter adhering in streaks or wide areas, making the surface appear fuscous (e.g. Bure with Coffee-color streaks in some young specimens, or Fox or Antique Brown), the whole varying from rather pallid to very dark; glutinous, smooth, glabrous, broadly campanulate-pulvinate or convex,

40–45 mm broad. *Hymenophore* tubulose, adnate, flat below, tubes ochraceous, 7.5 mm long, pores concolorous, up to 0.5 mm broad; spore-print cinnamon. *Stipe* whitish, finely gray-granular then ochre and soon showing brown, blackening glandules everywhere above and below the annulus; solid, eventually often somewhat hollow; usually tapering downward to an obtuse or acute base, 68–70 × 9–11 mm, rarely shorter; veil white, more rarely concolorous with the ground color of the pileus, often with glutinous, umbrinous-fuliginous streaks like the pileus on the outside, even occasionally blackening, connecting the margin of the pileus with the apex of the stipe where it forms a bag-like ring around the apex immediately beneath the pileus, later forming a characteristically sheathing, appressed annulus (the sheathing belt 10–20 mm broad), neither flaring nor ever forming a narrow zone in any individual but the sheath persisting and constant; basal tomentum sordid cream-color. *Context* a warm ochraceous-yellow (Amber Yellow) streaked and marbled a deeper orange especially in the stipe and mid-zone of the pileus, Gold Pheasant in the base of the stipe. *Spores* 8.3–9.2 × 2.7–3 μ, pale melleous; *basidia* 20–24 × 5.3–6 μ, 4-spored; *cystidia* pale melleous or lilac-colored, clavate, numerous, especially near the pores, 32.5–62 × 7.2–9 μ; *glandules* consisting solely of innumerable dermatocystidia up to 90 μ long and 7.3–10.5 μ broad, hyaline but with much lilac and pale melleous pigment present; *surface of the stipe* other than the glandules with some dermatocystidia and extremely few basidia, 20 × 5 μ; all *hyphae* without clamp-connections.

KOH on pileus surface Bone Brown (Bracken). NH_4OH on context of pileus dark violet-fuscous.

On poor soil especially around bogs, exclusively under two-needled pines, especially *Pinus resinosa* and *P. banksiana,* in small groups, fruiting in August. Formerly observed in northern and central Florida, now in northern Michigan from Luce County to Cheboygan County.

For the northern subspecies or race, we do not, for the time being, propose a new name, although the chances are that this is different from both the winter-fruiting and the summer-fruiting races of Florida. In the first place, we are not yet firmly convinced that the winter-fruiting race is different, and besides, there seems to be a chance that here also two races occur, based on their connection with either *Pinus resinosa* or *P. banksiana.* While the two races occur too often in mixed stands or too closely together, it seems to us that there is a possibility that the forms with whitish ground-color of the pileus are mycorrhizal with *P. banksiana,* and the other with *P. resinosa.* The evidence is, however, not conclusive. It is also not clear whether the color of the veil is in any way correlated with these two forms, and whether the conspicuously dark-colored individuals are always growing with *P. banksiana.* Further

observations here as well as elsewhere (Dr. Stuntz informs us that this species apparently occurs also in Washington) are necessary in order to complete an understanding of infraspecific taxonomy of this species.

Suillus tomentosus (Kauffm.) Singer, Snell & Dick, comb. nov.

Boletus tomentosus Kauffm., Pap. Mich. Acad. Sci., Arts & Letters **1**: 117. 1921.

Boletus hirtellus Pk. var. *mutans* Pk. ex Snell, Mycologia **33**: 26. 1941.

This species has been described as far as its remarkable macroscopic characters and its more important microscopic characters are concerned. We shall, therefore, confine ourselves to some information which has been neglected.

Pileus yellow, with red to olive-gray, appressed squamules which are tomentose and usually radially elongated except in the center of the pileus, distinctly and strongly viscid when wet, convex, becoming applanate, not umbonate, 50–78 mm broad. *Hymenophore* tubulose, slightly sinuate to adnate, 4.5 mm thick, 14–19 pores to 1 cm (measured in an arc at a distance halfway to the center), strongly bluing when fresh; spore-print deep olive. *Stipe* with light gray glandules which become deep purple or almost black. *Context* yellow, strongly bluing, often reddish in the cortical layer of stipe, more bright lemon-yellow in the pileus (much as in *S. Grevillei*), and strongly bluing when bruised. KOH on pileus deeper and more brownish, a weak reaction; on context brown. NH_4OH green to blue on context; deepening the color of the stipe, or making it more sordid.

On dry "islands" in a jack pine bog, i.e. around the Sphagnetum in a *Pinus banksiana* stand, sometimes near both *P. banksiana* and *P. resinosa*, but never under *P. resinosa* alone, apparently mycorrhizal with *P. banksiana*, in small to large groups, locally frequent, in August. Northern Michigan.

This collection of ample material is highly interesting because of its phytogeographical implications. *S. tomentosus* is a western species, particularly common from Washington to California, and seems to bypass the prairie belt in the north, reaching its easternmost limit in the United States in the area of Tahquamenon Falls State Park, Chippewa County. The exact color of the spore-print, the exact size of the pores, the characters of the surface of the pileus, and also the chemical reactions corroborate our earlier contention that this is a representative of the *Hirtellini* Sing. (subsection of *Suillus*), where it forms a stirps of bluing species together with *S. ruber* and *S. Cembrae*.

SUILLUS PIPERATUS

This species, otherwise typical, does not always occur in the neighborhood of pines. The carpophores are often rather large, at any rate well developed, and have a distinctly peppery taste, but they sometimes occur in frondose woods (mostly *Populus* and *Betula*) without any conifers present; only old stumps of *Tsuga canadensis* show that hemlock had been growing here years ago. While it would not be entirely unexpected to find *S. piperatus* not completely dependent upon the presence of conifers (in view of a similar situation in *S. rubinus* W. G. Smith, a closely related species), it is also possible that this species merely continues fruiting for a number of years after the conifers have been removed from its habitat. At any rate, this behavior is as atypical as that of *S. subaureus* in Michigan, and *S. rubinus* in England.

LECCINUM SCABRUM subsp. SCABRUM and L. OXYDABILE

These two taxa often offer certain difficulties in their distinction not only because *L. oxydabile* when old lacks the reddening of the context, but also because *L. scabrum* subsp. *scabrum* often does show a slight sordid pinkish discoloration on bruising. Furthermore, in northeastern Europe at least, Singer has encountered a variety (or subspecies?) of *L. scabrum* which turns strongly pink when young but is not identical with *L. oxydabile*, which he named var. *roseofractum* Sing. The following columns show the differences between *L. scabrum* subsp. *scabrum* and *L. oxydabile* as they occur in Cheboygan County, Michigan.

Leccinum scabrum subsp. *scabrum*	*Leccinum oxydabile*
Pileus smooth, somewhat *viscid* when wet, *tomentose, glabrescent,* color grayish-brown.	Pileus smooth or sometimes rugose, *not* distinctly viscid, *slightly velutinous* to *fibrillose,* not tomentose, grayish-brown to deep fuscous, eventually sometimes subglabrescent and mottled yellowish-brown to deep brown.
Pores whitish, on pressure not staining pinkish in the type variety.	Pores whitish, on pressure staining pinkish and finally brownish, only in very dry weather unchanging.
Scabrosities of stipe blackish.	Scabrosities of stipe a deep pure gray to black.

Context of pileus typically not turning pinkish by autoxidation, context of stipe white, in base sometimes yellow, not bluing in the white or yellow portions, and turning pink on bruising only occasionally in the apex of the stipe.	Context of pileus turning *pinkish on bruising* at least in the infrahypodermal zone, and that of the stipe, at least in the peripheral portion, also *frequently bluing* especially in the yellow portion of the base.
Cuticle of the pileus consisting exclusively of *filamentous* hyphae, with a diameter rarely over 4.5–6.2 μ (FIG. 1C).	Cuticle of pileus consisting partly of *broad* hyphae, the terminal and preterminal members of which are *frequently shortened* (28 × 15.5 μ, 19.5 × 13.5 μ, etc.), and even with some isolated sphaerocysts present in some specimens (FIG. 1D).
Formalin on context of pileus *pale pink* (Onion Skin Pink to 4B10).	Formalin on context of pileus *deep carrot red* (Carnelian or Copper) as in section *Decolorantes* of *Russula*, pink even in the lower portion of stipe.
$FeSO_4$ gray to bluish-gray in pileus (29E1 or grayer, sometimes more greenish-gray or Slate-Olive), negative in base of stipe.	$FeSO_4$ bluish-gray (29D-E1) to grayish-blue except in the base of the stipe.
Phenol on context slowly and moderately strongly copper-color, or almost negative.	Phenol on context chocolate-copper-color.

Both entities occur in similar habitats in Michigan. Where a strongly discoloring (pink) form of *L. scabrum* (var. *roseofractum*) occurs, one has to rely upon the main distinguishing character, the structure of the cuticle of the pileus. We have sectioned innumerable specimens from Asia and New England, and two from Michigan, and have discovered no transitions between the two.

The type of *L. oxydabile* was found in Central Asia (Teletskoye Ozero, Altay), by Singer and Vasiliev. Authentic material from both New England and Michigan is deposited at the Farlow Herbarium and the Chicago Natural History Museum.

The whitish, often greenish form of *L. scabrum*—subsp. *niveum*—is undoubtedly a hereditary race, occurring in or near *Sphagnum* in low woods here as well as in Europe. Whereas, however, most of the specimens of European collections, including the type of this as well as

Boletus holopus Rostk., are relatively smaller than the type subspecies, in eastern Europe, especially in Tartaria, and also in North America the whitish race has about the same size as the type subspecies. This is a *forma major*. It should not be confused with the white form of *L. aurantiacum*, which is considered to be a species by some European mycologists, but in view of intermediates, we believe this conclusion is erroneous. This white form of *L. aurantiacum* occurs also in Massachusetts and in Michigan along with a deep brown form (with no orange macroscopically) which is sometimes misinterpreted as *L. duriusculum* (Schulz.) Sing. by some European authors. The form of *L. aurantiacum* occurring under pines exclusively is also worthy of a distinguishing name since, in spite of the small number of characters distinguishing it, it seems to be a constant myco-ecotype, according to recent observations in Chippewa County, Michigan (material at Chicago Natural History Museum), and also in Finland, where it was reported by R. Tuomikoski [3] in an interesting account on Finnish representatives of the genus *Leccinum*. This pine form has grayish-white scabrosities which turn darker gray in age (instead of from white to dark reddish-brown), and the pileus has a peculiar deep brownish-red color which fades to sordid orange-brown in age. It can easily be confused with *L. testaceoscabrum* which occurs under *Betula*. This form has also been observed by Seeler and Snell in large quantities in the Cape Cod area of Massachusetts, where, upon Singer's insistence, a careful survey of the flora including the lower perennials was made, but no angiosperms likely to form mycorrhizae with Basidiomycetes were discovered. Seeler's specimens in particular were examined extremely carefully for possible anatomical differences from the type form of *L. aurantiacum* but no such differences were found. All of these observations and conclusions correspond closely with those of Tuomikoski.

III. SPECIES THAT MAY BE CONFUSED WITH XEROCOMUS CHRYSENTERON

Xerocomus truncatus Singer, Snell & Dick sp. nov.

A *Xerocomo chrysentero* praecipue sporis superne truncato-applanatis differt.

Pileus Tawny Olive to Isabella Color, or Buffy Brown to Olive Brown (all R), the cracks pinkish-purple or rarely with the pallid flesh showing through, dry, densely tomentose or velutinous, the fibrils tending to become agglutinated and aggregated into tufts, convex, expanding to plane or nearly so, 2–11 cm broad. *Hymenophore* depressed or sub-

[3] Tuomikoski, R. Notes on Finnish Agaricales. Karstenia **2**: 26-32. 1953 (pp. 27-28).

depressed around the stipe, Yellowish Citrine (R) but quickly changing to blue, readily separable from the context of the pileus; the tubes 10–15 mm long; the pores concolorous, rather large, about 1 mm broad, angular to irregular; spore-print olive. *Stipe* yellow at apex, otherwise red but with whitish to grayish-buff or tan-colored, cottony basal mycelium, or merely bister at base with little mycelial tomentum, with concolorous, fine, furfuraceous scurf almost all over or especially in the red mid-portion, sometimes with innate red striations, cylindric or slightly enlarged above or narrowed in the mid-portion; 4–10 cm × 8–20 mm. *Context* soft, pale yellow to whitish, at first quickly, later slowly, bluing upon bruising, the blue changing to pallid, the central portion of the stipe red; taste slightly acidulous; odor none.

Spores 12–17(–26) × (4.2–)4.5–6.5 μ, pale ochraceous-tawny in KOH, deep melleous in NH_4OH, always perfectly smooth, fusoid to subcylindric, with suprahilar depression or applanation, the mature ones often with rather firm wall, rarely with a spinulose lateral outgrowth, without germ-pore but with a truncate-applanate apex which is thicker-walled at the two apical angles and often with markedly thinned wall between these angles, there also frequently depressed to umbilicate-depressed, a minority with the apex merely rounded; in Melzer's reagent, pale tawny (not pseudo-amyloid or amyloid). *Basidia* 22–33 × 9–13 μ, clavate, hyaline to yellowish in KOH, 4-spored. *Cystidia* 32–70 × 8.5–12 μ, on pores and in tubes practically identical, fusoid to clavate-mucronate, the apices tapering to a subacute or almost obtuse narrow tip, pedicellate, the neck sometimes flexuous, hyaline to pale yellowish, numerous and projecting. *Subhymenium* hyaline, subcellular, of small elements; hymenophoral trama bilateral, the mediostratum not clearly delimited from the lateral stratum, the latter little differentiated except that it diverges, consisting of relatively large filamentous elements with numerous septa, 4.2–9.7 μ broad, nearly touching each other (i.e. relatively little separated by the mucilaginous mass which is not very abundant), the mediostratum consisting of hyaline and some golden, nearly parallel, slightly wavy hyphae with a diameter of 3–7.7 μ. All *hyphae* without clamp connections. *Covering layer* of pileus a trichodermium consisting of a palisade of hyphae pale ochraceous in KOH, with the terminal members parallel or nearly so, erect or eventually sometimes depressed-horizontal, either elongated and 32–93 × 8.7–16.3 μ, or rarely with a terminal sphaerocyst, the tips broadly rounded, pigment-encrusted, pale ochre to ochraceous-brown but thin and often scanty.

Chemical characters not studied.

In mixed woods, often among mosses, fruiting in summer and fall.

Material studied: **CANADA** — Quebec, Mt. Burnet, *F. L. Drayton 20*, July 21, 1935 (MICH, DAOM); Ontario, Calabogie, *B. P. Tomlin*, Aug. 22, 1955 (DAOM). **U.S.A.** — North Carolina, Highlands, *W. C.*

Coker 12426, TYPE, *12395,* July 26, 1941 (UNC, FH); Washington, Mt. Baker National Forest, *A. H. Smith 16277,* August 9, 1941 (MICH); Mt. Rainier National Park, *Stuntz & Simmons,* A. H. Smith 30769, Aug. 29, 1948 (MICH).

The description given above shows clearly that, in spite of the extraordinary shape of the apex of the spores, this species has the hymenophoral structure of a *Xerocomus* (*Phylloporus*-type) rather than that of a *Boletellus.* Consequently, it is here treated as a *Xerocomus* closely allied to *X. chrysenteron.* A new name has been applied, since Coker & Beers's determination as *Boletus chrysenteron* is most certainly erroneous, because a species with spores like those exhibited by this species is not known to occur in Europe, whereas the species we consider as the true *X. chrysenteron* is common in Sweden as well as in the remainder of Europe.

The two species are macroscopically very similar, and in many cases it is necessary to examine the spore-shape in order to arrive at a safe determination. On the other hand, the shape of the apex of all or the majority of the spores is so striking that a single glance at a random preparation from the hymenophore will immediately decide the issue.

Boletellus Zelleri (Murr.) Singer, Snell & Dick, comb. nov.

Ceriomyces Zelleri Murr., Mycologia **4**: 99. 1912.
Boletus Zelleri Murr., Mycologia **4**: 217. 1912.
Xerocomus Zelleri (Murr.) Snell apud Slipp and Snell, Lloydia **7**: 43. 1944.

Pileus Prussian Red to Seal Brown or Cameo Brown (R), with the margin somewhat paler and more brown, appearing glabrous when wet but when dry plainly pruinose with a very delicate, pallid bloom which disappears with age, often with a submetallic lustre, firm, convex to subapplanate, up to 18 cm broad. *Hymenophore* pale grayish-yellow to olivaceous-yellow, unchanging, adnate-depressed or slightly decurrent; pores concolorous, rotund to subangular, about 2 a mm; spore-print ochraceous-brown. *Stipe* at first entirely light to dark pinkish-red, becoming Chamois (R) and obscurely red-streaked below with the apex remaining red, at the base white to chalky-yellow, more or less striate or grammate, furfuraceous, perhaps glabrescent, at base matted-tomentose or coactate, subequal or subbulbous, perhaps flexuous, 5–9 cm × 7–15 mm. *Context* of the pileus pallid or cremeous, pinkish-red beneath the cuticle, unchanging or becoming more or less blue after very long exposure, firm, but light and dry, in the stipe whitish to pale yellow in the center, red to the outside, firm and tough, fibrous with a carti-

laginous cortex, unchanging; odor none; taste mild and slightly mucilaginous.

Spores 11–17.5 × 4.8–6.5 μ, fusoid-oblong, in profile fusoid-cylindric, with a suprahilar depression, apparently smooth but with scarcely elevated, extremely faint and sometimes intermittent, longitudinal striation (visible best in phase microscope in diluted Melzer solution near the lower, hilar end of the spore), golden-melleous or melleous-citrine, with moderately thin wall. *Basidia* 26–35 × 9.5–12 μ, hyaline in KOH, 4-spored. *Cystidia* 38–77 × 5.5–14.8 μ, numerous, practically identical on edge and inside the tubes, at first cylindric to subcylindric, later becoming enlarged below or in the middle, ampullaceous with an obtuse apex about 5 μ broad which remains cylindric or more rarely without such apex, hyaline or subhyaline or more rarely brownish, thin-walled. *Hymenophoral trama* of the *Boletus*-subtype (general in *Boletellus*), not of the *Phylloporus*-subtype (as in *Xerocomus*), the lateral stratum in young specimens strongly gelatinized, strongly divergent and consisting mostly of relatively very narrow hyphae which are often wavy in the gelatinous mass in which they are imbedded. All *hyphae* without clamp connections. *Epicutis* of the pileus a trichodermium but the trichodermium arranged in individual tufts and erect strands of deep chestnut-colored hyphae; the terminal cells chestnut to hyaline and frequently cystidioid, these dermatocystidia 18–35 × 4–14 μ, with thin to moderately thin wall, sometimes with a median septum, versiform (fusoid-ventricose, cylindric or cylindric-flexuous, clavate, capitate, ampullaceous, filamentous, subulate, etc.), with a subacute to broadly rounded tip.

Chemical characters not known.

Mainly in mixed coniferous forests on the humus, more rarely on decayed wood, in mountain forests, fruiting from July until November.

Material studied: **CANADA** — British Columbia, Stanley Park, Vancouver, *Mary Alice Wough 47*, Oct. 31, 1949, det. Singer (DAOM, MICH); **U.S.A.** — Washington, Nisqually River, 2300 ft. elevation, *Imshaug 2130*, Aug. 30, 1948, det. A. H. Smith (MICH); Oregon, TYPE (NY); Mt. Hood, *C. H. Kauffman*, Oct. 7, 1922, det. W. A. Murrill, authentic (MICH); Mt. Tabor, *W. B. Gruber*, Nov. 14, 1942, det. Singer (MICH, also a stereophoto); Portland, *W. B. Gruber*, Nov. 22, 1942, det. Singer (MICH); and many other collections from the same general region (MICH, Herb. WHS).

The ornamentation of the spores is difficult to observe because it is faint but its presence is sufficiently constant to be of more than incidental interest with regard to spore-ornamentation as a definitive character in the Strobilomycetaceae. The ornamented spores of this species are indeed accompanied by a distinctly bilateral trama of the *Boletus*-subtype in young specimens, although in older specimens the hyphae of

the lateral trama become stretched rather strongly and appear so nearly subparallel that one would interpret them as belonging to the *Phylloporus*-subtype, at least to the degree seen in *Xerocomus badius*. The correlation of the *Boletus*-subtype hymenophoral-trama structure with ornamented spores makes it impossible to leave this species in *Xerocomus* in spite of the deceptive external similarity to *X. chrysenteron*. In reality, the tubes are long and sufficiently depressed to demonstrate a *Boletellus* habit, and the surface of the pileus is similar to that found in certain other species of that genus.

With this transfer, we may reopen the question of delimitation of the genus *Boletellus* versus the Boletaceae. Some specialists have been confused by the presence of species with smooth spores in the genus *Boletellus*, because, whereas in *Strobilomyces* and *Porphyrellus* smooth spores can still be readily identified as being strobilomycetaceous even if smooth, in *Boletellus* this was not necessarily the case because of the color and shape of many of the spores involved.

It is therefore interesting to note that, of those species that constitute the *chrysenteron* complex (not stirps!), one with a development alien to *Xerocomus* is now shown to have ornamented spores. On the other hand, two smooth-spored species tentatively inserted in *Boletellus* by Singer (see footnote 1, I, p. 134), viz. *Boletus turbinatus* Snell and *B. chrysenteron* sensu Coker (now the new species *truncatus* above), are actually upon detailed re-examination demonstrably species of other genera—the former of *Gastroboletus*, the latter of *Xerocomus*. This leaves only one smooth-spored species in *Boletellus*, *B. mirabilis* (Murr.) Sing. This species has extraordinarily large spores a certain percentage of which, according to recent investigations by A. H. Smith, are vaguely ornamented, although the percentage is even lower than in *B. Zelleri* and the roughening even less apparent.

These observations contribute to a better delimitation of *Boletellus* from other bolete genera, particularly from *Xerocomus*. They also confirm the importance of the spore-ornamentation as a family character in the Strobilomycetaceae, and of the *Phylloporus*-subtype of hymenophoral trama as a character peculiar to the Xerocomoideae.

BROWN UNIVERSITY
PROVIDENCE, R. I.
AND
INSTITUTO MIGUEL LILLO
TUCUMÁN, ARGENTINA

NEW AND INTERESTING SPECIES OF BASIDIOMYCETES VII [1]

ROLF SINGER

(WITH 13 FIGURES)

PSILOCYBE LONGINQUA Sing. spec. nov.

Pileo aurantiaco-luteo, ad marginem obscuriore (aquose balteato?), 20 mm lato. Lamellis cremeis, decurrentibus. Stipite cremeo-flavo, evelato, basin versus attenuato, 25 × 10 mm. Sporis 8.8–11 × 6.5–8.8 μ, depressione applanationeque suprahilaribus destitutis. Tramate hymenophorali subregulari. Cystidiis hyalinis, gradatim densius dispositis aciem versus, tenui-tunicatis, hyalinis, ventricoso-subfusoideis, 32–53 × 7.5–14.5 μ. Hyphis epicuticularibus plus minusve gelatinascentibus, hypodermio vix manifeste subcellulari.

Pileus orange yellow, darker at margin (with darker watery belt?), glabrous, smooth, without traces of veil, obviously viscid or subviscid when wet, with incurved margin, umbonate, 20 mm broad.

Lamellae cream-color, not anastomosing, decurrent.

Stipe cream yellow, evelate, fistulose, tapering downward from a broad apex, 25 × 10 mm.

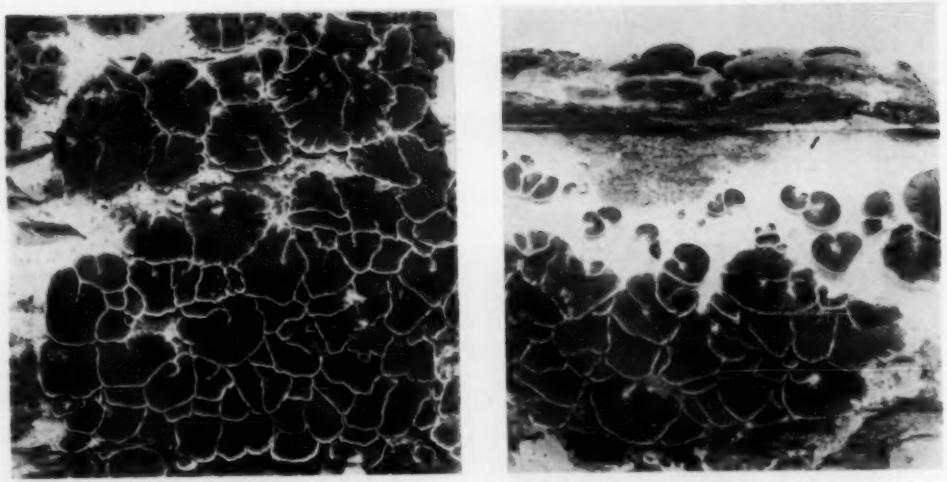
Context pallid (either white or cream), firm, neither soft nor fragile in pileus and gill trama, thick in pileus, cartilaginous-fleshy in stipe. Odor and taste not recorded.

Spores, 8.8–11 × 6.5–8.8 μ, ellipsoid, without suprahilar depression or applanation, with a narrow not flattened germ pore, ochraceous brown in ammonia but dusky reddish fuscous in KOH, smooth, with triple wall.

Hymenium: Basidia 27.5–29 × 8.8–9.7 μ, clavate, 4-spored. Cystidia 32–53 × 7.5–14.5 μ, hyaline, crowded on edge, and progressively more scattered towards the flesh of the pileus, ventricose-subfusoid, usually broadest in the middle and tapering above and below, obtuse at apex, thin-walled, but often seemingly thick-walled (because of plasma bridges and plasma condensations in the upper portion along the wall and consequently often appearing chambered) in alcohol-formalin material, often with constrictions, often with appendiculations like those of the *Russula* macrocystidia, especially in the cheilocystidia.

Hyphae with clamp connections. Hymenophoral trama subregular, i.e., of somewhat interwoven filamentous elements, pale yellowish. Hy-

[1] A continuation of the paper published under this title in Mycologia 51: 375–400. 1959.



FIGS. 1, 2. *Crepidotus harperi* Sing. Photographs of the type, in fresh condition, phot. Harper; copies kindly authorized by the Director, Natural History Museum, Chicago.

podermium not distinctly subcellular. Epicutis consisting of thin undulant, distant filamentous hyphae (in KOH).

On humus on north (sun-exposed) slopes of the plateau at 800 ft elevation. In late fall.

Australian Subantarctic: Macquarie Island, 1–V–1949, *Bunt*, TYPE (LIL).

This differs from *Naematoloma laeticolor* in broader spores.

Psilocybe squarrosipes Sing. spec. nov. FIG. 3

Pileo flavobrunneo-subcinnamomeo, margine subochraceo, vel subalbo, centro interdum subtiliter scruposo vel squamuloso, ceterum levi, viscido, 20–24 mm lato. Lamellis purpureo-griseis dein rubello-fuscis, latis, subconfertis vel subdistantibus, late adnexis; sporis in cumulo obscure ferrugineo-sepiaceis vel sepiaceo-fuscis. Stipite albo, fortiter albo-fibrilloso-floccoso, 34–37 × 3–4 mm. Carne alba, inodora, miti. Sporis 12–12.7 × 7.5–8 μ. Cystidiis nullis; cheilocystidiis cylindraceis 33–43 × 5–5.5 μ. Epicute plus minusve gelatinosa, ex hyphis filamentosis repentibus efformata; hypodermio vix subcellulari.

Pileus yellowish brown tending toward light cinnamon (12–J–9, M&P), on margin golden ochraceous *daffodil* (10–J–6, M&P) to nearly white, smooth except sometimes slightly uneven to subsquamulose in the center, viscid, convex, more rarely conic-applanate, acutely or obtusely umbonate, 20–24 mm broad. Veil appendiculate or marginal on the surface of the margin, pure white, floccose, denticulate.

Lamellae purple gray, then chocolate, with pallid edges, subclose to (more rarely) subdistant, broadly adnexed. Spore print deep rusty sepia to sepia-fuscous.

Stipe white, strongly white fibrillose-floccose-squarrose, tending to be or become concolorous with the margin of the pileus, at least in the basal third of the stipe, the latter often entirely buried in the substratum, hollow, equal or subequal, 34–37 × 3–4 mm. Veil not definitely annular although there is a circular line where the veil breaks off from the stipe to join (in youth) the marginal veil.

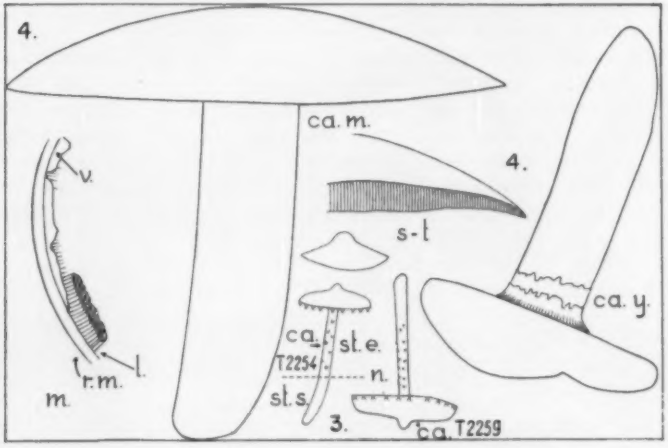
Context white, or whitish, including the base of the stipe except in very old material (and then there concolorous with the surface), unchanging, fleshy. Odor none. Taste mild.

Spores 12–12.7 × 7.5–8 μ, ellipsoid, with strongly developed broad germ pore, smooth, deep rusty both in NH_4OH and KOH.

Hymenium: Basidia 28–32 × 10–11.3 μ, 4-spored. Cystidia on sides of lamellae none. Cheilocystidia numerous, 33–43 × 5–5.5 μ, with very thin wall above, and moderately thin wall at base, cylindrical, with or without a slight constriction in the middle, sometimes subcapitate, hyaline.

Hyphae with clamp connections. Hymenophoral trama regular, hyaline to palest ochraceous.

Covering layer of pileus: Epicutis somewhat gelatinized, consisting of repent hyphae forming a cutis, these hyphae filamentous, hyaline or subhyaline, not incrusted, all thin and thin-walled, hypodermium also cutis-like and consisting of filamentous hyphae but these much denser



FIGS. 3, 4. FIG. 3. *Psilocybe squarrosipes* Sing. ca. carpophores; st.e. epigeous portion of stipe; st.s. subterranean portion of stipe; n. level of soil surface, × 1. FIG. 4. *Suillus lithocarpi-sequoiae* Sing. ca.m. mature specimen of type collection (stipe somewhat shortened); ca.y. younger specimen of type collection; s-t. section of pileus showing tubes and margin; m. margin of pileus seen from below showing rim of margin (r.m.), appendiculate veil (v.), and lamellar zone of hymenophore (l.), all × 1.

and not quite as regularly arranged as the hyphae of epicutis, also mostly with distinct rusty ochraceous membrana pigment, which incrusts some of the hyphae, elements not subisodiametric.

Solitary and in small groups on alpine pastures among *Stipa*, sometimes growing on sandy soil containing a certain amount of dung, and sometimes directly on deep *Stipa* turf.

Argentina: Prov. Tucumán, Infiernillo, 10/11–III–1955, *Singer T 2254*, HOLOTYPE, and *T 2259* (LIL).

A characteristic species of the alpine zone growing between 2900 and 3200 m alt. in the Aconquija Mountains (Pre-Andine Sierras).

Psilocybe pteridophytorum Sing. spec. nov.

Pileo obtuso, 5 mm lato. Lamellis subdistantibus. Stipite dactylicolori pileo pallidiore, velo nullo ornato, 20 × 1 mm. Sporis 7.8–8 × 4.7–5.2 × 4.3–4.7 μ, paulum lentiformibus; cheilocystidiis exiguis, centro ventricosis, cystidiis facialibus nullis.

Pileus deep dark brown, glabrous, subviscid, short and indistinctly striate, not umbonate, convex, about 5 mm broad; veil white, on the extreme margin, not striking.

Lamellae brownish, subdistant, broad, adnate.

Stipe date brown, paler than the pileus, about equal, about 20 × 1 mm. Veil on stipe none, at least when mature.

Context partly dull brownish, without any striking odor.

Spores 7.8–8 × 4.7–5.2 μ (in frontal view), vaguely rhomboid (benzene formula), when turned around their longitudinal axis and seen in profile, they are ellipsoid and 4.3–3.7 μ broad, i.e. remarkably little broader in frontal than in lateral view although still visibly lentiform (about as much as in *Psilocybe albofimbriata* (Rick) from Brazil), melleous brown, smooth, with broad germ pore, thick and complex walled.

Hymenium: Basidia 4-spored. Facial cystidia, none seen. Cheilocystidia numerous, 25–30 × 4.7–5.3 μ, i.e. small, ventricose in the midportion, acute or subacute at the tip, but often with a round mucilaginous head which is very fugacious, hyaline, apical part of cheilocystidium 1–2 μ in diameter.

Hyphae of the gill trama brownish, of the epicutis repent.

On fallen fern thallus in subtropical mountain forest.

Argentina: Prov. Tucumán, Rio de los Sosas, 4–II–1955, *Singer T 2132*, TYPE (LIL).

This banal-looking little species is apparently well separated from related species of the same group, i.e. *Deconica*. This group was formerly separated from *Psilocybe* on the basis of spore size (small) and shape (lentiform), absence of cystidia, and chemical inactivity of the context. Numerous species studied carefully during the last years have turned out to be transitional in the sense that now not one of the diagnostic characters of *Deconica* is characteristic for this genus alone, while the combination of the characters enumerated above is not apt to circumscribe a natural group separated by a recognizable hiatus. Consequently, we have already (Mycologia **50**: 246 (footnote). 1958) mentioned that A. H. Smith, who does not recognize the genus *Deconica*, is probably right, and in the present paper the new deconicas (this and the following species) are described as *Psilocybe*.

We also propose the following new combination:

Psilocybe alnetorum (Sing. *in* Sing. & Digilio, Lilloa **25**: 332. (1951), 1952, ut *Deconica*) Sing. comb. nov. FIG. 13

Psilocybe peruviana Sing. spec. nov. FIG. 14

Pileo ferrugineo-alutaceo striolato, glabro, convexo, centro depresso, velo nullo visibili statu maturo, 5–7 mm lato. Lamellis melleo-argillaceis, adnatis, subdistantibus, latiusculis. Stipite alutaceo, sericeo, evelato, 13 × 1 mm. Sporis 6.7–7.8 × 5.6–6 × 4.4–5 μ, lentiformibus, cystidiis ad latera lamellarum sparsis vesiculosis, cheilocystidiisque hyalinis, his ventricosis 25–27 × 5.2–6.5 μ, epicute haud gelatinisata.

Pileus over one half of the radius ferrugineous buff transparently striate on melleous buff ground, not viscid, glabrous, smooth, or, when dried, sulculate, convex with depressed center, not papillate, 5–7 mm broad.

Lamellae melleous argillaceous, rather broad, subdistant, adnate.

Stipe buff, slightly pallid, longitudinally silky, equal or subequal, 13 × 1 mm. Veil none, at least not in mature specimens.

Context thin, inodorous.

Spores 6.7–7.8(–9) × (frontally) 5.6–6(–7.2) × (profile) 4.4–5 μ, olive melleous (dusky), smooth, lentiform, with distinct truncate germ pore, with complex thick wall.

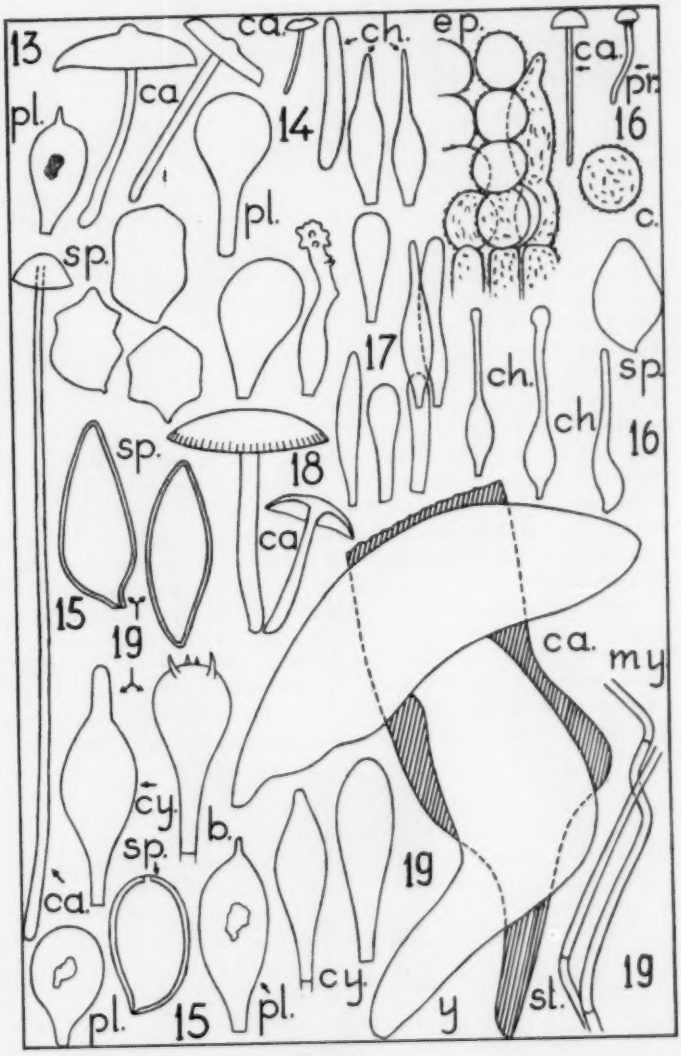
Hymenium: Basidia 22–26 × 7.5–8.2 μ, clavate, hyaline in NH_4OH and KOH, 4-spored. Cystidia 16–32 × 10–13.5 μ, vesiculose or almost vesiculose, hyaline, without yellow or hyaline amorphous enclosures, thin-walled, not very numerous and less so toward the trama of the pileus, more so toward edge. Cheilocystidia differentiated, making the edge heteromorphous, hyaline, ampullaceous, ventricose in mid-portion or below, with acute or obtuse tip, not incrusted, 25–27 × 5.2–6.5 μ.

Hyphae with clamp connections, rather pale in the context and hypodermium of the pileus where they are often short and subvesiculose, making up a subcellular layer but not sharply separated from the tissue of the pileus, upper zone of hypodermium with distinct incrusting ochraceous brown pigment, same pigment incrustation also in the cutis of the epicuticular layer but only in its lower layer while the uppermost layer of the pileus is hyaline or subhyaline, not gelatinized; all elements of the epicutis filamentous and thin-walled.

In small moss carpets under scrubs in semi-xerophytic conditions fruiting in fall.

Peru: Dpto. Cuzco, Lamay, 3000 m alt., 21–V–1958, *Singer B 3032*, TYPE (LIL).

This new species is microscopically and ecologically rather well characterized. It grew in a small group below the high shading rocks but above the fertile valley. Macroscopically it is banal and not striking in any way.



FIGS. 13–19.[2]

[2] Numbers continue those of the sixth paper of this series.

Pholiota glutinigera Sing. spec. nov. FIG. 15

Pileo aureo-brunneolo. Stipite glutinoso, elongato. Sporis in cumulo eis *Pholiotae aurivellae* concoloribus, 11.3–13.3 × 7–7.5 μ, poro germinativo lato praeditis. Chrysocystidiis numerosis; cheilocystidiis hyalinis. In silvis montanis.

Pileus olive brown to gold brown (marginal zone *gold*, 14–K–17, M&P), short transparently striatulate to distinctly striate over 4/5 of the radius, convex, hygrophanous, becoming ochraceous yellow in faded condition, viscid, 8–16 mm broad, mostly about 12 mm in diameter.

Lamellae argillaceous to olive, pallid fringed eventually rusty brown, subclose to subdistant, adnexed or more often adnate, not ventricose. Spore-print color was compared with that of *Pholiota aurivella* and appears identical.

Stipe pallid whitish to greenish lemon color at apex, fulvous to yellow or concolorous with marginal portion of pileus in the lower part, pallid pruinate at apex, glutinous up to a sharply delimited circular zone, but not distinctly annulate or even often without any sign of a veil, tapering upward, hollow, 70–110 × 1–1.5 mm (at apex), base 2.5–5 mm thick.

Context pallid but partly colored like the surfaces, thin, not very fragile. Odor not strong, practically none.

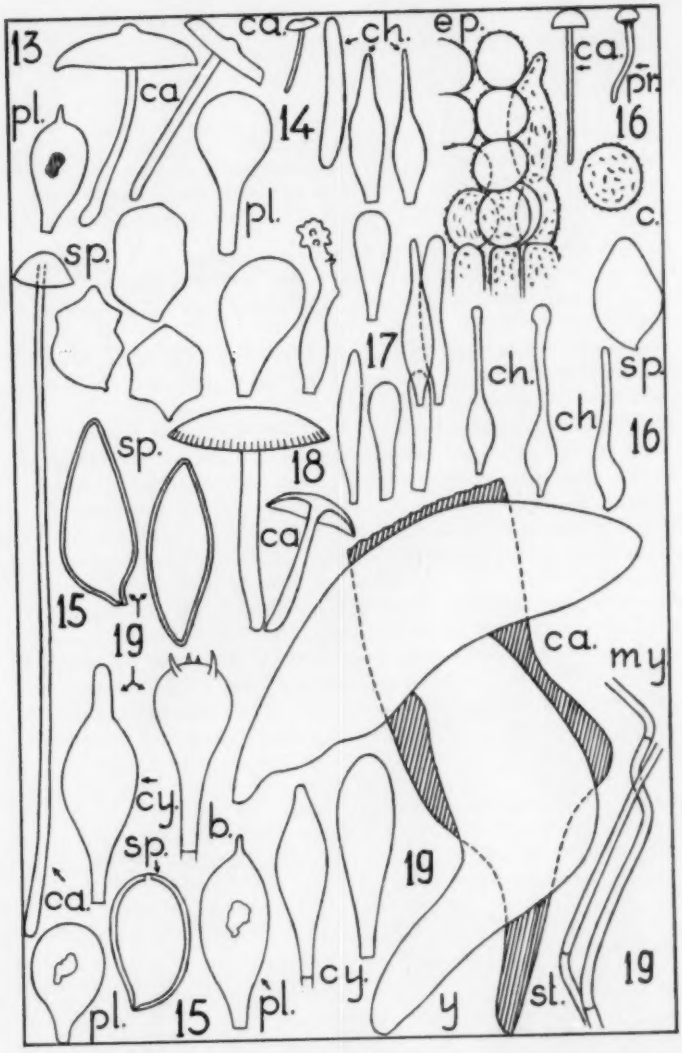
Spores 11.3–13.3 × 7–7.5 μ, rusty ocher brown, ellipsoid, smooth, with thick, double wall, with broadly truncate germ pore.

Hymenium: Basidia 22.5 × 8.7 μ, 2- or 4-spored, normally mostly 4-spored, very few 3-spored. Chrysocystidia rather numerous on the 2 sides of the lamellae, 14.5–34 × 10.7–13.5 μ, with light yellow amorphous enclosure (in KOH), clavate-vesiculose, with or without an acute or obtuse appendage or mucro. Cheilocystidia entirely hyaline and without enclosures, 42–64 × 6.7–8.7 μ, "empty" or with a very fine granulosity, almost all subulate, very few subcylindric, mostly strongly projecting.

Hyphae all with clamp connections. Subhymenium irregular and somewhat intermixed, consisting of some subisodiametric elements, mixed with some more elongated ones which are interwoven to interlaced but

---

FIGS. 13–19. FIG. 13. *Psilocybe alnetorum* Sing. pl. pleurocystidium; ca. carpophores. FIG. 14. *Psilocybe peruviana* Sing. pl. pleurocystidia; ca. carpophore; ch. cheilocystidia. FIG. 15. *Pholiota glutinigera* Sing. ca. carpophore; sp. spore; pl. pleuro-(chryso-)cystidium. FIG. 16. *Phaeomarasmius gregarius* Sing. ep. epicuticular layer; c. an isolated epicuticular spherocyst; ca. carpophore; pr. primordial carpophore; sp. spore in profile; ch. cheilocystidia. FIG. 17. *Crepidotus harperi* Sing. cheilocystidia. FIG. 18. *Acurtis sericeus* (Bull. ex Fr.) ssp. *antarcticus* Sing. sp. spores; ca. carpophores. FIG. 19. *Pulveroboletus flaviporus* (Earle) Sing. sp. spores; cy. cystidia; b. basidium; cast. stipe of adult specimen; y. carpophore of a young specimen; my. hyphae of basal mycelium. (All spores × 2000; other microscopical drawing × 1000; macroscopical drawing × 1.)



FIGS. 13–19.[2]

[2] Numbers continue those of the sixth paper of this series.

Pholiota glutinigera Sing. spec. nov. FIG. 15

Pileo aureo-brunneolo. Stipite glutinoso, elongato. Sporis in cumulo eis *Pholiotae aurivellae* concoloribus, 11.3–13.3 × 7–7.5 μ, poro germinativo lato praeditis. Chrysocystidiis numerosis; cheilocystidiis hyalinis. In silvis montanis.

Pileus olive brown to gold brown (marginal zone *gold*, 14–K–17, M&P), short transparently striatulate to distinctly striate over 4/5 of the radius, convex, hygrophanous, becoming ochraceous yellow in faded condition, viscid, 8–16 mm broad, mostly about 12 mm in diameter.

Lamellae argillaceous to olive, pallid fringed eventually rusty brown, subclose to subdistant, adnexed or more often adnate, not ventricose. Spore-print color was compared with that of *Pholiota aurivella* and appears identical.

Stipe pallid whitish to greenish lemon color at apex, fulvous to yellow or concolorous with marginal portion of pileus in the lower part, pallid pruinate at apex, glutinous up to a sharply delimited circular zone, but not distinctly annulate or even often without any sign of a veil, tapering upward, hollow, 70–110 × 1–1.5 mm (at apex), base 2.5–5 mm thick.

Context pallid but partly colored like the surfaces, thin, not very fragile. Odor not strong, practically none.

Spores 11.3–13.3 × 7–7.5 μ, rusty ocher brown, ellipsoid, smooth, with thick, double wall, with broadly truncate germ pore.

Hymenium: Basidia 22.5 × 8.7 μ, 2- or 4-spored, normally mostly 4-spored, very few 3-spored. Chrysocystidia rather numerous on the 2 sides of the lamellae, 14.5–34 × 10.7–13.5 μ, with light yellow amorphous enclosure (in KOH), clavate-vesiculose, with or without an acute or obtuse appendage or mucro. Cheilocystidia entirely hyaline and without enclosures, 42–64 × 6.7–8.7 μ, "empty" or with a very fine granulosity, almost all subulate, very few subcylindric, mostly strongly projecting.

Hyphae all with clamp connections. Subhymenium irregular and somewhat intermixed, consisting of some subisodiametric elements, mixed with some more elongated ones which are interwoven to interlaced but

---

FIGS. 13–19. FIG. 13. *Psilocybe alnetorum* Sing. pl. pleurocystidium; ca. carpophores. FIG. 14. *Psilocybe peruviana* Sing. pl. pleurocystidia; ca. carpophore; ch. cheilocystidia. FIG. 15. *Pholiota glutinigera* Sing. ca. carpophore; sp. spore; pl. pleuro-(chryso-)cystidium. FIG. 16. *Phaeomarasmius gregarius* Sing. ep. epicuticular layer; c. an isolated epicuticular spherocyst; ca. carpophore; pr. primordial carpophore; sp. spore in profile; ch. cheilocystidia. FIG. 17. *Crepidotus harperi* Sing. cheilocystidia. FIG. 18. *Acurtis sericeus* (Bull. ex Fr.) ssp. *antarcticus* Sing. sp. spores; ca. carpophores. FIG. 19. *Pulveroboletus flaviporus* (Earle) Sing. sp. spores; cy. cystidia; b. basidium; cast. stipe of adult specimen; y. carpophore of a young specimen; my. hyphae of basal mycelium. (All spores × 2000; other microscopical drawing × 1000; macroscopical drawing × 1.)

show some axillar arrangement, deeper colored than the trama proper. Hymenophoral trama subregular, i.e., regular but interwoven, consisting of mostly remarkably large (and sometimes short, even subisodiametric) elements, melleous-hyaline, the short elements e. g. 27 × 18.8 μ. Trama of the pileus rather interlaced, melleous-hyaline.

Covering layer of pileus: Epicutis consisting of more or less gelatinized, repent filamentous hyphae which become less gelatinized as they approach the hypodermium, many elements beset with an incrusting ocher brown to rusty pigment, mostly those closer to the hypodermium. The latter a cutis consisting of densely packed strictly thin-filamentous hyphae (not subcellular), not gelatinized, pigment-incrusted like the lower layer of the epicutis but even more strongly so.

In montane woods on the ground on detritus, rotting fern leaves, etc., under *Sambucus*, *Alnus*, and *Podocarpus*, solitary but not uncommon in its zone (1200–1900 m altitude, east slope of Andes and Pre-Andes).

Argentina: Prov. Tucumán, Taficillo (Sierra de San Javier), 4–III–1951, *Singer T 1336* (LIL), 23–II–1955, *Singer T 2217*, TYPE (LIL). Las Languas, 20–II–1951, *Singer T 1284* (LIL). Also numerous other collections and observations in the upper zone of the Selva Boliviana-Tucumana.

This species is remarkable since it is a perfect replica of the *Stropharia semiglobata* group within the limits of the genus *Pholiota*, but reminding one rather of some of the glutinous naematolomas as far as habit is concerned. This is the type of a group of *Pholiota* which deserves to be placed in a section all by itself. I propose:

Sectio **Glutinigerae** Sing. sect. nov. Stipite glutinoso. TYPUS sectionis: *P. glutinigera* Sing.

Phaeomarasmius gregarius Sing., Schweiz. Zeitschr. Pilzk. **34**(4): 58. 1956, nom. subnud., spec. nov. FIG. 16

Sporis pallide stramineis, 8.2–9.9 × 5.1–6.2 μ, subfusiformi-ellipsoideis. Pileo argillaceo (*clay*, 13-J-8, M&P), margine subpallidiore haud pellucido. Cheilocystidiis 29–34 × 4.8–5.5 μ, ampullaceis. Elementis epicuticularibus pilei elongatis trichodermium formantibus. Ad terram humidam viae silvestris. *Singer V 302*, TYPE (FH).

A more detailed description has been published in Schweiz. Zeitschr. Pilzk., l.c.

Crepidotus harperi Sing. spec. nov. FIGS. 1, 2, 17

Quam *C. applanatus* minor, sporis minoribus, superficie pilei tomentosa, mycelio extenso; quam *C. quitensis* maior, mycelio extenso nec strigoso, pileo mycelioque tomentosis albis.

Pileus white, tomentose, convex then repand, smooth, on horizontal surfaces pendulous-applicate on lower side, on vertical surfaces broadly attached in the rear and extending horizontally, 7–19 mm broad.

Lamellae whitish then brown as in most *Crepidoti*, about 12 through-lamellae present, but strongly intermixed (regularly or almost so), narrow to medium broad, never broad (i.e. lamellae never much more than $\frac{1}{10}$ of pileus diameter in breadth), close, rounded-concurrent at stipe or the place where it was when carpophore was younger.

Stipe rudimentary, white, extremely small and disappearing at maturity, functionless; basal mycelium covering the entire substratum, white, tomentose.

Context white, thin. Taste and odor not recorded.

Spores 4.3–5.7 μ diam., globose, mostly between 4.7 and 5.3 μ large, with deeply inserted cylindric spinules which are deeper ferruginous than the melleous ground color of the episporial material in which they are imbedded, punctate when seen from above and with the upper surface of the spores in focus.

Hymenium: Basidia 13–15 × 4.7–5.7 μ, 2-, 3-, or mostly 4-spored. Cystidia none. Cheilocystidia 17–30 × 3.5–7.5 μ, versiform, but frequently cylindric-irregular, also clavate or subulate, with obtuse tip, sometimes nodulose like the cheilocystidia of certain *Marasmielli*, hyaline, thin-walled.

Hyphae of the gill trama and base of basidium usually clamped. Mycelium with or without clamp connections, in some hyphae alternating with clampless septum and clamped septum, most hyphae slightly thick-walled with the protoplasmatic contents concentrated near the septa.

On wood; densely gregarious, fruiting in spring.

U.S.A.: Virginia, Bedford, May 1919, *Harper 1177*, HOLOTYPE, *1178* (F).

Acurtis sericeus (Bull. ex Fr., Epicr. 147. 1838, ut *Agaricus*) Sing., comb. nov., ssp. **antarcticus** Sing., ssp. nov. FIG. 18

A ssp. *sericeo* differt sporis 8.2–9 × 6.2–7 μ stipiteque breviore.

Pileus of a medium dark brown, finely striate on margin, smooth, convex, 20–30 mm broad.

Lamellae medium dark brown, moderately broad, close, adnate.

Stipe light brown below, dark brown above, fistulose, 20–30 × 3 mm.

Spores 8.2–9 × 6.2–7 μ, angular, pinkish-stramineous, without basal tetrahedra (i.e. axially asymmetric).

Hymenium: Basidia 35–36 × 8.5–9 μ, clavate, 4-spored. Cystidia none, but there are in some specimens occasional basidiomorphous bodies with a cylindric mucro instead of sterigmata.

Hyphae and bases of basidia mostly but not all with clamp connections. Hymenophoral trama regular, consisting of filamentous hyphae, fuscidulous, individual hyphae, however, with little pigmentation, frequently with granular covering.

Covering layer of pileus not very well individualized, the whole cuticular zone being a cutis consisting of more or less parallel hyphae which are often incrusted by pigment as in the omphalinas, and besides with colored or hyaline contents.

On humus in a *Juncus* swamp, summer fruiting, apparently locally common.

Australian Subantarctic: Macquarie Islands, Half Moon Bay, 31–I–1951, leg. *Bunt*, TYPE (LIL). Also from other localities (hills above Green Gorge, Sawyer Creek, Green Gorge).

This belongs in the group *Nolanidei* as delimited by Kühner & Romagnesi. It keys out with what these authors call "var. *typicus*" but differs in slightly narrower spores and relatively shorter stipe.

GOMPHIDIUS SUPERIORENSIS Kauffm. & Smith, Pap. Mich. Acad. Sci. **17**: 170. 1933

When writing my monograph of the North American species of *Gomphidius*, I had not had occasion to see fresh material of this apparently rather rare species. Good specimens were found, however, during August 1953, in Chippewa Co., northern Michigan, and it was established that this *Gomphidius* is definitely connected by mycorrhiza with *Pinus resinosa* while *G. vinicolor* ssp. *vinicolor* took up the habitats under *Pinus banksiana*, and *G. septentrionalis* those under *Pinus strobus*, *Picea mariana*, etc. The impression from the fresh material is that it differs rather strongly from *G. ochraceus* in its colors, and it may be preferable to maintain it as an autonomous species rather than a subspecies of *G. ochraceus*. In this case, the eastern *G. ochraceus* ssp. *muscigenus mihi* becomes a subspecies of *G. superiorensis* rather than of *G. ochraceus*.

The color of the mycelial tomentum at the base of the stipe is a very good distinctive character in this group of *Gomphidii*. It was observed that it has to be used on dried material. The color of the basal tomentum in *G. superiorensis* is not distinctive, as far as the *G. rutilus* complex is concerned, but becomes pinkish to sordid pink as soon as the specimen is dried, revealing its affinity with *G. ochraceus*.

GOMPHIDIUS SEPTENTRIONALIS Sing., Pap. Mich. Acad. Sci. **32**: 149. 1948.

Specimens of this species were found in the same neighborhood where *G. vinicolor* and *G. superiorensis* were encountered. This gave occasion to a revision of its chemical characters. The entire salmon-colored portion of the pileus, context and cuticle become deep pink in NH_3 vapors and in ammonia water (NH_4OH); in the yellow portion of the flesh, NH_4OH provokes an almost rufous discoloration after a while. The entire context, including the deep chrome-yellow base, blackens strongly with $FeSO_4$. The color of the pileus was found to be *Longchamps* to *monkey skin* of M & P. The yellow part of the stipe is 9-L-5 to *Florida gold*, M & P, i.e., deeper colored than in *G. glutinosus*. The carpophores were up to 50 mm broad, and the stipe up to 53 × 12 mm, altogether little different from the type specimen. The only trees found around these carpophores were consistently *Pinus strobus*, *Picea mariana*, and *Betula*. This species is new for Michigan.

Suillus lithocarpi-sequoiae Sing. spec. nov. FIG. 4

Pileo glabro, glutinoso. Tubulis decurrentibus. Velo glutinoso. Stipite glandulis destituto, solido. Carne caerulescente. Sporis 7.5–9.8 × 3.2–3.8 μ. Hyphis defibulatis. In silva *Sequoiarum* prope *Lithocarpos*.

Pileus cinnamon, in some places at times buffish, or reddish brown to cinnamon buff (rarely uniformly colored), glutinous all over in young and old specimens, more or less rugulose, drying rapidly, convex, obtuse, 70–140 mm broad: margin entire, acute.

Hymenophore tubulose; tubes golden yellow to almost primuline yellow, later dingy olive ocher to dusky olive brown, with small brown dots, not very long (up to 8 mm long), rather short decurrent, arcuate, then flat below; pores concolorous with the tubes, subisodiametric but not strictly circular or boletinoid-elongated, only in a zone very close to the margin of the pileus radially elongated and sublamellar, eventually slightly boletinoid even in inner (rear) portion and becoming compound and many of them over 1 mm in diameter. Tube walls linear-decurrent on the extreme apex of the stipe. Spore print not obtained.

Stipe either pallid to stramineous or buffish pallid to pale brownish, ferruginous squamulose or ferruginous furfuraceous above and below annular belt but only within a zone of 10 mm in both directions, or particles becoming very scattered as the middle of the stipe is approached, otherwise subglabrous to glabrous, solid, equal except for the basal fifth to third which is attenuated downwards but not acute or radicant, 80–110 × 22–33 mm; basal mycelium white; veil entirely glutinous (a thick sordid mucilaginous mass) on a membranous whitish or buff-

colored fundamental tissue, forming an apical superior annulus which is appressed, often lacerate to denticulate on both margins, not distant, merely oblique and belt-like; volva none.

Context whitish or buffish white, bluing moderately quickly and moderately deeply, the blue stains soon becoming fuscous, the discolorations more intense in the stipe, often browning directly (without first becoming blue) in some parts, in old specimens more lemon yellow all over, and less discoloring, firm but soon soft. Taste mild. Odor agreeable, weak, fruity, somewhat like that of *Boletinus cavipes*.

Spores 7.5–9.8 × 3.2–3.8 μ, cylindric-subfusoid to oblong, smooth, with rather thin wall, pale melleous to melleous-stramineous.

Hymenium: Basidia 29–31 × 6–7 μ, elongate-clavate, hyaline, 4-spored. Cystidia numerous on pores and slightly less numerous inside the tubes, single or more often in fascicles, rarely hyaline, usually with a brown resinous, broken or continuous incrustation which dissolves vinaceous in KOH or is accompanied by a vinaceous (in KOH mounts) soluble pigment especially in younger specimens, cylindrical or somewhat ventricose, 48–84 × 4–10.5 μ.

Hyphae all without clamp connections, subhymenium (like sometimes also the basidioles) hyaline and becoming dyed by the vinaceous pigment of the incrustations of the hymenial cells (in KOH), consisting of irregular small elements. Hymenophoral trama bilateral of the *Boletus* subtype; lateral stratum quite hyaline, divergent, its hyphae not touching each other, separated by a gelatinous mass; mediostratum pale melleous to subhyaline, consisting of axial slightly interwoven hyphal elements.

Covering layer of pileus consisting of a pellicle of thin, wavy to straight, hyaline to yellow or melleous hyphae which are sometimes incrusted, and always imbedded in a gelatinous mass, less distant in the outermost layer where they are also more incrusted than further inward, and also less distant in the innermost layer, which is very weakly differentiated from the trama of the pileus, all hyphae of the covering layer characteristically narrow, 1–2.2 μ in diameter. Incrustations melleous (in flesh more frequently hyaline), strongly dyed blue in cresyl-blue mounts.

On the ground in mixed woods (*Sequoia* and *Lithocarpus*) in small groups. Fruiting in winter.

U.S.A.: California, Marin County, Muir Woods, 30–I–1958, *Singer N 1531*, TYPE (LIL).

It may justly be asked why this species is not rather a *Boletinus* than a *Suillus*. The glabrous pileus which is glutinous in all parts, even in youth, the not very distinctly boletinoid hymenophore, and the strongly incrusted cystidia seem to be in favor of *Suillus*. On the other hand,

the species is rather isolated in *Suillus* (as it would likewise be in *Boletinus*), inasmuch as it has some characters which are unusual in both genera, such as the arcuate-decurrent hymenophore of the young carpophores, the lamellate marginal zone (both again encountered in the related Asiatic genus *Psiloboletinus*), and the unusual habitat.

Since we were unable to obtain more than an extremely thin spore layer, we do not wish to propose a new section for this species at the present time, although we are strongly convinced that it deserves to be separated in a special section near the section *Larigni*. It is also desirable to obtain more definite data on the mycorrhizal status of this species. It is very probable that it is mycorrhiza-forming, as are all species of its subfamily, but we are unable to state whether this is one of few (or the only known?) species of the Basidiomycetes forming mycorrhiza with redwood, or whether, like *Suillus rubinus*, it is connected with a representative of the Fagales.

It may also be asked whether the natural relationship of this species, especially in case its mycorrhiza, if any, should be with hardwoods, is not rather with *Pulveroboletus* in the sense of the present author than with the Suilloideae. In the first place, however, this latter genus would have to be emended if *Suillus lithocarpi-sequoiae* were transferred to it, and in the second place, the color of the spore print, not known at present, should be examined before such a transfer is seriously considered.

PULVEROBOLETUS FLAVIPORUS (Earle) Sing., Am. Midl. Natur. **37**: 17. 1947 FIGS. 5, 6, 19

Boletus flaviporus Earle, Bull. N. Y. Bot. Gard. **3**: 297. 1905.

Pileus chestnut-reticulate on pale tan ground or entirely chestnut or cinnamon-chestnut color, viscid, eventually sometimes rivulose, pulvinate, 85–120 mm broad; veil pallid pelliculose-tomentose in youth, appendiculate on margin.

Hymenophore tubular. Tubes golden yellow, then deep golden sulphur yellow, depressed around stipe, ventricose, long; pores concolorous, between "light cadmium" and "anilin yellow" (R.) when dried, becoming dusky gray-olive on pressure or when bruised, eventually brown-dotted, gyrose, reaching more than 1 mm in diameter at maturity, extended on the extreme apex of the stipe and running down it over a distance of approximately 5 mm, in thin lines.

Stipe concolorous with pileus, mostly chestnut-cinnamon, or (in youth) yellow punctate on yellow to yellowish white ground, more cinnamon below, more or less ribbed-rugose (longitudinally), the elevated parts yellow fibrillose, otherwise glabrous, with a smooth and glabrous subacute or acute projecting and deeply buried pseudorhiza, solid, thick

FIGS. 5, 6. *Pulveroboletus flaviporus* (Earle) Sing. FIG. 5. Carpophores in natural size, topotypes, phot. R. Singer.

FIG. 6. Carpophores in natural size, topotypes, phot. R. Singer.

and cylindric or gradually attenuated towards the apex but the base always strongly tapering downwards into the earth, sometimes abruptly so, 35–50 (70–85 with pseudorhiza) × 25–36 mm (at apex 15–30 mm broad); annulus none; volva none; basal mycelium white.

Context white, unchanging, firm, eventually very soft, especially in the pileus. Taste mild. Odor slight, insignificant.

Microscopical characters see Singer, Am. Midl. Nat. l.c.

Chemical characters: NH_4OH on young pores, green; on older pores, brown; on other organs, negative.

On earth in groups under oak. Fruiting in winter.

U.S.A.: California, Santa Clara Co., Stanford University grounds, *C. F. Baker,* Nov. 11, 1901 (Pacific Slope Fungi no. 131, FH, TYPE).—*R. Singer N 1575,* 7–II–1958 (MICH, LIL, TOPOTYPE).

Since the species had not been re-collected (or at least not been reported as having been re-collected) since it was first discovered and distributed by Baker, it seems useful to publish a revised description, inasmuch as some of the characters constant in this bolete have not been brought out in the type description. This is particularly true regarding the veil.

INSTITUTO MIGUEL LILLO
TUCUMAN, ARGENTINA

NOTES AND BRIEF ARTICLES

Some Physiological Properties of Geotrichum candidum [1]

Geotrichum candidum and similar fungi are frequently encountered in dairy products and the respiratory and digestive tracts of man and other warm-blooded animals. For this reason the group has been of special interest to dairy microbiologists and medical mycologists. There are still points of controversy as to how far these fungi are active in certain dairy processes, such as the ripening of Camembert cheese, and whether they may produce disease (geotrichosis) in warm-blooded animals. In connection with this controversy sometimes the question is raised whether strains which are commonly called *G. candidum* (or for which one of the more common synonyms, like *Oidium lactis,* is used) really belong to one single species, or if some of them belong to other species which, among other distinctive properties, might have special dairy-fermentation capacity or pathogenicity towards animals. Names like *Oidium camemberti* and *Geotrichum pulmoneum* reflect this way of thinking.

Recently it was shown in this journal by Carmichael [2] that the species *Geotrichum candidum* can be recognized by morphological means and that the great majority of *Geotrichum* strains isolated from dairy, human and animal sources belong to this species.

Windisch,[3] studying physiological as well as morphological properties, decided at an earlier date in favor of the taxonomic identity of most strains looking like *G. candidum.*

Completing and extending Carmichael's and Windisch's work, we studied a number of physiological properties in 23 strains, which by the use of Carmichael's morphological criteria had to be identified with *G. candidum.*

Origin of strains: Intestinal tract of humans (2 strains), of cattle (3 strains), of horses (2 strains), of rabbits (2 strains), and of a sheep (1 strain); a swine embryo (1 strain); Camembert cheese (1 strain);

[1] Part of this work was subsidized by the Direcção Geral dos Servicos Pecuários, Ministry of Agriculture, Lisbon, and by the Instituto de Alta Cultura, Lisbon.

[2] Carmichael, J. W. *Geotrichum candidum.* Mycologia **49**: 820-830. 1957.

[3] Windisch, S. Zur Biologie und Systematik des Milchschimmels und einiger ähnlicher Formen. Beit. Biol. Pflanz. **28**: 69-130. 1951.

Roquefort cheese (1 strain), Kefir grains (2 strains); yogurt (2 strains); unknown (6 strains).

Fermentation (according to Wickerham's [4] technique): All strains (23) form acid in glucose broth and all but one form gas. Gas formation is slow: 16 strains form just a gas bubble, 6 others fill most or the whole of the insert with gas after a minimum of from 7 to 15 days. In galactose broth acid is formed by 20 of the 23 strains; by 4 of these acid-forming strains also a gas bubble is formed. Neither acid nor gas is formed in sucrose, maltose, or lactose broth.

Assimilation (according to Wickerham's technique): All strains use glucose, galactose, L-sorbose, D-xylose, ethyl alcohol, glycerol, D-mannitol, and D-sorbitol. No strain uses any of the following compounds: sucrose, maltose, lactose, cellobiose, trehalose, melibiose, raffinose, inulin, soluble starch, L-arabinose, L-rhamnose, i-erythritol, dulcitol, i-inositol, potassium nitrate.

Maximum temperature for growth: The strain was grown for 24 hours at 25° C in broth of glucose (2%), peptone (1%), and yeast extract (0.5%). A loopful of the suspended growth was then inoculated into a tube with 5 ml of the same medium and the tube was incubated in a water bath at constant temperature. Growth was recorded daily for five days. In this way the maximum temperatures of 19 strains were determined. As can be seen from FIG. 1, the maximum varies between 35° and 38°; the graph suggests a logarithmic function.

Vitamin requirements (according to the technic of van Uden et al.; [5] incubation at 25°): All strains but two grow well in the absence of external vitamins. The two deficient strains (isolated from the intestinal tract of a rabbit and a horse, respectively) require pyridoxine for growth.

Fat splitting (according to the technic of Eijkman, as described by Lodder & Kreger-van Rij [6]): Positive reactions were obtained with all strains within a few days.

Discussion: Twenty-three strains of various origins, including human and animal faeces as well as dairy products, which by the use of Carmichael's morphological criteria had to be classified as *Geotrichum candidum*, showed a high degree of uniformity in their physiological properties. All strains behaved identically in assimilation tests with

[4] Wickerham, L. J. Taxonomy of yeasts. Tech. Bull. U. S. Dept. Agr. no. 1029. 1951.

[5] Uden, N. van, L. do Carmo Sousa and M. Farinha. On the intestinal yeast flora of horses, sheep, goats and swine. Jour. Gen. Microbiol. **19**: 435–445. 1958.

[6] Lodder, J., and N. J. W. Kreger-van Rij. The yeasts, p. 33. Amsterdam. North-Holland Publishing Company. 1952.

23 different compounds. Windisch observed slight variation among his strains with regard to assimilative behavior toward 10 different compounds; possibly this is due to his using an auxonographic method with a solid medium, which is less sensitive than the method of Wickerham with liquid media used by us. All our strains split fat. Only two of the strains were deficient for a single vitamin, the others being independent.

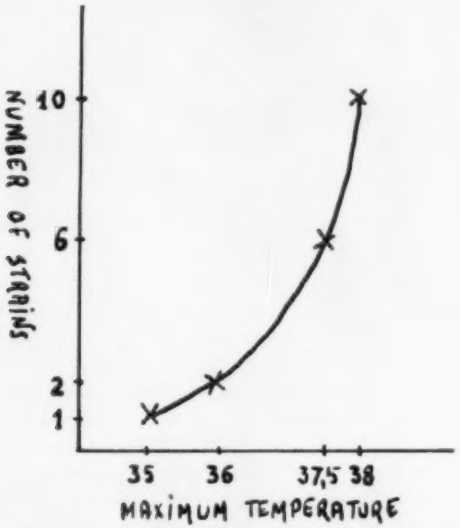


FIG. 1. Distribution of maximum temperatures for growth among 19 strains of *Geotrichum candidum*.

Some variation was observed with regard to acid and gas formation in glucose and galactose broth; this, however, may be of a quantitative nature only, as is suggested by the fact that all strains used glucose and galactose in the assimilation tests.

From the curve showing the distribution of the maximum temperature among the strains it can be learned that a number of strains are not able to grow at the body temperature of man and certain animals, whereas other strains are. Certainly, therefore, not every strain of *Geotrichum candidum* is a potential pathogen; this naturally does not mean that the strains with the higher maximum temperature should necessarily be potential pathogens.—N. VAN UDEN AND LIDIA DO

CARMO-SOUSA, Department of Microbiology, Botanical Institute, University of Lisbon, Lisbon, Portugal.

STEMONITIS BRASILIENSIS AND BADHAMIA IOWENSIS—A CORRECTION

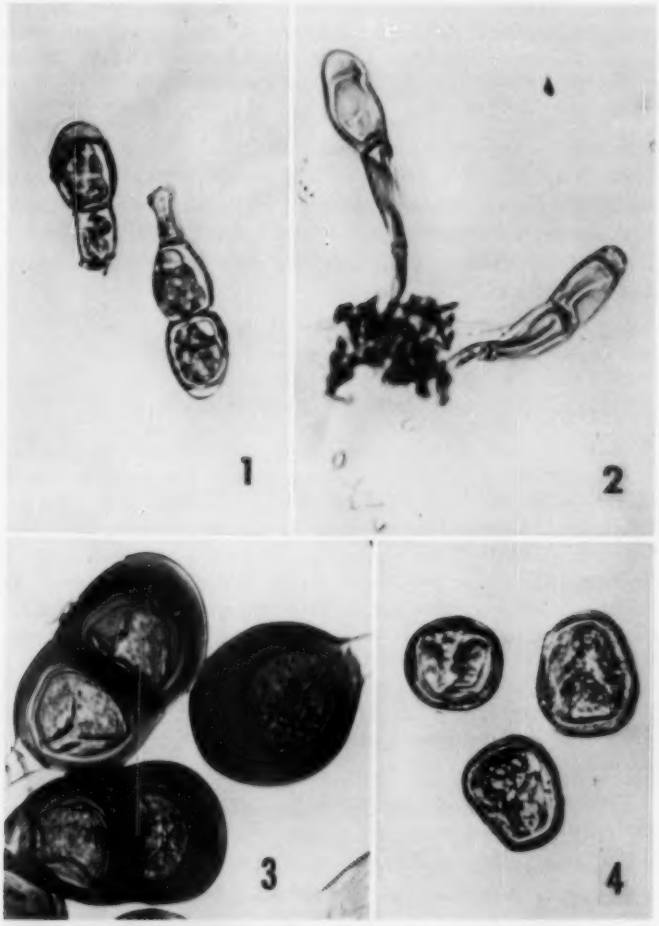
In a recent publication (Farr, Marie L., & George W. Martin, 1958. Brotéria Ciênc. Nat. **27**(4) : 156) we described a new species of *Stemonitis, S. brasiliensis,* from Brazil. Shortly thereafter, I had occasion to reexamine a report of Indian Myxomycetes containing an illustrated diagnosis of a new species, *Stemonitis mussooriensis* Martin, Thind, & Sohi, Mycologia **49**: 128. 1957. The descriptions and pictures of these two organisms indicated that they might be one and the same species, and a comparative study of type material of both confirmed this suspicion. Since these slime molds are apparently identical, the earlier name, *S. mussooriensis* Martin, Thind, & Sohi, must take precedence according to the International Code of Botanical Nomenclature, and *S. brasiliensis* Farr & Martin becomes a synonym.

During a study of collections of *Physarum auriscalpium* Cke. a portion of the type of *Badhamia iowensis* Macbr. was discovered. The latter has been considered a pale phase of *P. auriscalpium* by Lister (Mycetozoa 3 ed., p. 40. 1925) and G. W. Martin (N. Am. Fl. **1**(1) : 107. 1949) and listed by both as a synonym of that species. Macroscopic and microscopic examination of this fine specimen demonstrate it to be in all respects typical *Physarum pusillum* (Berk. & Curt.) G. List. The pure-white color of the lime on the grayish peridia and in the capillitium nowhere displays the slightest trace of yellow (as it does in faded specimens of *P. auriscalpium*). The spores have scattered, more prominent warts or wart clusters in addition to the minute over-all wart pattern, an occurrence not mentioned in the description, but indicated in Lister's illustration. Thus "*Badhamia iowensis* Macbr." should be listed as a synonym of *Physarum pusillum* (Berk. & Curt.) G. List.—MARIE L. FARR, Crops Research Division, Agricultural Research Service, U. S. Department of Agriculture, Beltsville, Maryland.

NOTES ON LABIATE RUSTS

During a study of the rusts occurring on the Labiatae in North America, specimens of *Monarda didyma* L. and *Monarda media* Willd. were found to bear telia of a previously undescribed microcyclic rust, which is here described as a new species. This paper also presents

amplified descriptions of two previously described rusts on *Salvia, Puccinia mellifera* Diet. & Holw. and *Puccinia farinacea* Long var. *azurea* Baxter. Specimens examined during this study were obtained through the courtesy of Dr. G. B. Cummins, Purdue University, Dr. D. P.



FIGS. 1, 2. Teliospores of *Puccinia monardae* (from type) × 800. FIG. 3. Teliospores of *Puccinia mellifera* (from type) × 800. FIG. 4. Aeciospores of *Puccinia mellifera* (from type) × 800.

Rogers, University of Illinois and Dr. H. C. Greene, University of Wisconsin.

Puccinia monardae sp. nov. FIGS. 1, 2

Spermagoniis non visis, verisimiliter nullis. Aeciis et urediis nullis. Teliis plerumque hypophyllis, 0.5–1.0 mm diam., flavidis vel pallide cinnamomeo-brunneis, compactis. Teliosporis clavatis vel oblongo-ellipsoideis, 10–14 × 23–46 μ, ad apicem rotundatis vel truncatis, deorsum rotundatis vel contractis, ad septum constrictis; membrana 0.5–1.0 μ, ad apicem 3–7 μ, hyalina, levi; pedicello fragili, 5–20 μ longo.

SPECIMENS EXAMINED: On *Monarda didyma* L., Cheat Bridge, West Virginia, Aug. 18, 1906, *John L. Sheldon 2434*, TYPE. On *Monarda media* Willd., Ronceverte, W. Va., Sept. 25, 1904, *John L. Sheldon*.

This species resembles *Puccinia delicatula* (Arth.) Sacc. & Trott., but differs in the length of the pedicel and the pronounced apical thickening of the teliospore wall.

PUCCINIA MELLIFERA Diet. & Holw. in Diet., Erythea **1**: 25. 1893. FIGS. 3, 4

Allodus mellifera Arth., Res. Sci. Congr. Bot. Vienne 345. 1906.

Spermagonia not seen. Aecia chiefly hypophyllous, in groups or scattered, cupulate or cylindric, with conspicuous peridium; aeciospores angularly globoid, ellipsoid or oblong, 15–25 × 22–33 μ; wall 2–4 μ thick, cinnamon-brown or yellowish, finely striate-verrucose, pores visible, scattered, capped by yellowish or hyaline cuticle. Uredia lacking. Telia amphigenous and caulicolous, 0.5–1.5 mm in diameter, scattered, compact, blackish-brown or chocolate-brown; teliospores ellipsoid, oblong or subclavate, 23–31 × 35–48 μ, rounded at the apex, rounded or somewhat narrowed at the base, constricted at the septum; wall 3–6 μ at the sides, thickened to 7–9 μ at the apex, chestnut-brown, smooth; pedicel persistent, thick-walled, yellowish next to the spore, hyaline downward, 30–120 μ long. Mesospores globoid, 21–35 μ in diameter, wall 3–7 μ thick.

SPECIMENS EXAMINED: On *Salvia sonomensis* Greene, Montgomery Creek, Calif., June 12, 1923, *E. Bethel*. On *Salvia mellifera* Greene, Pasadena, Calif., 1893, *McClatchie*, TYPE.

This species resembles *Puccinia bithynica* Magn., a *Salvia* rust of the Mediterranean region.

PUCCINIA FARINACEA Long var. AZUREA Baxter in Baxter & Cummins, Lloydia **14**: 221. 1951.

Urediospores broadly ellipsoid or obovoid, 17–23 × 20–25 μ, or globoid to oblate-spheroid, 18–25 μ broad by 16–21 μ high; wall 1–2 μ, cinnamon-brown, echinulate, pores 2 or 3, subequatorial or occasionally equatorial. Teliospores ellipsoid or oblong, 18–22 × 24–35 μ, wall about 2 μ thick, thickened to 5–7 μ over the pores, chestnut-brown, moderately to obscurely verrucose, occasionally appearing smooth.

SPECIMENS EXAMINED: On *Salvia azurea* Lam., Lincoln, Neb., Sept. 1887 (ILL 21147). On *Salvia reflexa* Hornem., Albany, Green Co., Wis., *H. C. Greene.*

This variety is intermediate between the typical *Puccinia farinacea* and *Puccinia caulicola* Tr. & Gall.—JOHN W. BAXTER, University of Wisconsin—Milwaukee.

REVIEWS

MICHAEL/HENNIG HANDBUCH FÜR PILZFREUNDE; Erster Band, Die wichtigsten und haüfigsten Pilze mit besonderer Berucksichtigung der Giftpilze, revised by Bruno Hennig. viii + 260 pp., 17 figs., 120 colored pl., 2 color charts. Gustav Fischer, Jena, 1958. Price, 36.50 DM bound.

This volume is the first of four in a new treatment of Michael's "Führer für Pilzfreunde." As proposed by Hennig, the present editor, the set is to include the descriptions and colored illustrations of about 1000 fungi. The text is in German. Volume 2 is to include the fleshy and woody fungi other than the agarics; volume 3 includes the light-spored agarics; volume 4 includes the dark-spored agarics.

Volume 1 is designed primarily for popular appeal and contains 200 fleshy and woody fungi which the author considers to be the most common and important. Most of the species are known in North America and the book should be of considerable use to the amateur. The first 128 pages consist of such topics of general information as: poisons and poisonings, characteristics of the various types of carpophores, chemical reactions used in the study of fungi, the classification of fungi, collecting and preserving fungi. Of unusual interest in this introductory portion is an extensive list of the odors found in agarics and other fleshy forms.

The principal value for most mycologists will be the 200 descriptions and illustrations. There are no keys to genera or species, and some students will find this a disadvantage. On the other hand, a particularly convenient feature is that the description and illustration of a species are always on facing pages. The fungi are adequately described in regard to field characteristics and to their edible or poisonous qualities. The descriptions do lack complete information on all microscopic features of the carpophore and usually only the size and shape of spores are given. The plates are not consistent from one to another in the quality of color and execution of the drawings. Certainly the majority are good representations of the intended species and are quite usable for determinations. However, several artists prepared the paintings and individual styles are evident. Some plates fail to be a realistic rendition.

The good features of this first volume seem to outweigh the shortcomings, and the completed set should be a significant contribution to the study of higher fungi.—HOWARD E. BIGELOW.

NAŠE HOUBY II. KRITICKÉ DRUHY NAŠICH HUB. (OUR MUSHROOMS. II.), by A. Pilát and O. Ušak. 345 pp., 160 colored plates. Czechoslovakia Academy of Sciences, Prague, 1959. Price, 100 Czechoslovakian crowns (about $12.00).

Dr. Pilát and his artist associate, the late O. Ušak, present a second volume on the fleshy fungi. The first volume, which appeared in Czech, German, and English editions some six years ago, described and illustrated with 120 colored plates a series of the more common edible and poisonous mushrooms of Europe. In this second volume with 160 colored plates less known or critical species are considered. The plates are of the same high degree of excellence as those in the first volume. Each species is reproduced natural size or approximately so and in different stages of growth and coloration; this will facilitate to a marked degree the use of the plates as direct aids in species identification. The genus *Cortinarius* is emphasized by providing for 46 selected species the adequate illustrations so indispensable for exact identifications in this genus, which is very difficult because of critical variations in color and other ephemeral characters. Many other genera of fleshy fungi, particularly *Boletus* and its segregates, *Tricholoma, Russula,* and *Lactarius* are also included.

The text accompanying each plate includes an adequate description with notes on the occurrence, distribution and the edible or poisonous qualities of each species. Critical synonymy is cited and comparative data provided where necessary to distinguish related species. Because of the artistic and technical excellence of the plates, the book will be not only of interest but of decided usefulness even to those who are not familiar with the language employed. It is to be hoped, however, that an English edition will be made available.—JOHN A. STEVENSON.

DIAGNOSTIC DE LABORATOIRE EN MYCOLOGIE MEDICALE, by G. Segretain, E. Drouhet, and F. Mariat. 143 pp., 25 figs. Editions de la Tourelle, Saint-Mande, Seine. 1958. Price, 930 Fr.[1]

One of the outstanding European laboratories devoted to research and teaching in the field of medical mycology is the Service de Mycologie of the Pasteur Institute in Paris. Dr. G. Segretain, director of the Service, and his colleagues Drs. E. Drouhet and F. Mariat have

[1] From the Communicable Disease Center, Public Health Service, Department of Health, Education, and Welfare, Atlanta, Georgia.

made many important contributions towards a deeper understanding of the various mycoses and their etiologic agents.

The publication of a manual describing the diagnostic procedures currently in use at the Pasteur Institute should be an event of unusual interest to workers in this field. The reader will find it profitable to examine the techniques, media and tests that have been tried and evaluated by the authors.

The material is organized into 7 chapters. The first one briefly reviews some of the basic properties of fungi and serves to orient the student embarking upon diagnostic work. In the next chapter the various types of diseases produced by human pathogenic fungi are described and the basic methods used to establish a diagnosis are outlined. Specific procedures for the microscopic examination of clinical materials and methods for isolating and identifying the pathogens are presented in chapter 3. The next sections furnish detailed instructions for the laboratory diagnosis of the cutaneous, subcutaneous and systemic mycoses. In the 7th and final chapter formulae for mounting and clearing fluids, stains, culture media, slide-culture procedures, sealing compounds and indicators are provided. The text is illustrated with 25 simple outline drawings. A glossary that defines 34 specialized terms is appended along with a short bibliography of pertinent reference books. A table of contents is provided but no index.

On the whole, this description of the methods currently in use at the Pasteur Institute is a handy summary of modern techniques. However, the laboratory worker lacking experience with fungi and mycological procedures will find it necessary to supplement this text with other publications that furnish complete descriptions as well as detailed photographs of the cultural and histological aspects of the pathogenic fungi.

Several statements and some of the nomenclature employed by the authors are disputed: *Trichophyton verrucosum* is the validated name for the trichophytons of the *faviforme* group (*T. album*, *T. discoides* and *T. ochraceum*); *T. schoenleinii* is not exclusively a human parasite, since several well authenticated lower-animal infections have been described by Catanei and others; the binomial *T. megninii* is considered by many mycologists to have priority over *T. rosaceum* and the fungus is not known to infect chickens; mucormycosis is caused by several species of *Rhizopus* as well as members of the genus *Mucor*.

The format of this booklet is pleasing. Its size, although small, makes it handy to use and the type employed is highly legible.—LIBERO AJELLO.

SUSTAINING MEMBERS
OF THE MYCOLOGICAL SOCIETY OF AMERICA

The Society is grateful for the support of its Sustaining Members. They are listed below in alphabetical order.

ABBOTT LABORATORIES
Pharmaceutical products for the medical profession since 1888
North Chicago, Illinois

BALTIMORE BIOLOGICAL LABORATORY, INC.
A division of Becton, Dickinson and Company
Products for the microbiological laboratory

BUCKMAN LABORATORIES, INC.
Industrial microorganism control specialists
Memphis 8, Tennessee

BUTLER COUNTY MUSHROOM FARM
Incorporated
West Winfield, Pennsylvania

CIBA PHARMACEUTICAL PRODUCTS INC.
In the service of medical science
Summit, New Jersey

DIFCO LABORATORY PRODUCTS
The complete line of
microbiological reagents and media

THE FLEISCHMANN LABORATORIES
Standard Brands Incorporated

JOHNSON & JOHNSON
New Brunswick, New Jersey

LEDERLE LABORATORIES DIVISION, AMERICAN CYANAMID COMPANY
Antibiotic, biological and pharmaceutical research and manufacture
Pearl River, New York

ELI LILLY AND COMPANY
Pharmaceuticals, biologicals, and agricultural and industrial products
740 South Alabama Street, Indianapolis, Indiana

MILES LABORATORIES, INCORPORATED
Pharmaceutical and chemical research and manufacture
Elkhart, Indiana

NORWICH PHARMACAL COMPANY, EATON LABORATORIES DIVISION
Ethical pharmaceuticals
Norwich, New York

(Continued)

SUSTAINING MEMBERS OF THE MYCOLOGICAL SOCIETY OF AMERICA

(Continued)

ORTHO PHARMACEUTICAL CORPORATION
Obstetrical & gynecological products, biologicals
Raritan, New Jersey

CHAS. PFIZER & CO., INC.
11 Bartlett Street, Brooklyn 6, New York
Fine chemicals and pharmaceuticals by means of microorganisms

SCHERING CORPORATION
Pharmaceutical manufacturers
Bloomfield, New Jersey

THE SQUIBB INSTITUTE FOR MEDICAL RESEARCH
E. R. Squibb & Sons, Div. of Olin Mathieson Chemical Corporation
New Brunswick, New Jersey

ARTHUR H. THOMAS COMPANY
22,000 items of chemical and biological apparatus for immediate shipment
Vine Street at Third, Philadelphia, Pennsylvania

THE UPJOHN COMPANY
Fine pharmaceuticals since 1886
Kalamazoo, Michigan

MANUSCRIPT

Publication in MYCOLOGIA is ordinarily restricted to those who have been members in good standing of the Mycological Society of America for over a year immediately preceding submission of manuscript. Exceptions to this regulation require a favorable vote by a majority of the Editorial Board. When a paper has two or more authors, the person submitting the paper is expected to be a member.

Papers should be submitted in duplicate, typewritten and *double-spaced throughout*, to any member of the Editorial Board. When papers are not submitted in proper form, it may be necessary to return them to authors. They will be published in approximate order of their acceptance, except for the address of the retiring President and papers whose cost of publication is paid by the authors, the latter run as excess pagination.

All illustrations should be numbered consecutively throughout a paper, using arabic numbers and small letters for subdivisions, e.g., Fig. 1, a etc. This does not mean that all figures grouped for convenience on a single page need have a single number. Figures should be prepared so that, when reduced, the width will not exceed 4 inches, and should be short enough to permit the insertion of the legend beneath the figures. Each article will be restricted to twenty pages, including illustrations, except when authors submit only one paper in two or three years of membership, in which case the restriction will be thirty and forty pages respectively. Ruled tabular matter is counted double. Should an author wish to publish additional pages in one article he may do so by paying for the excess pages at current rates.

Citations of literature should be *double-spaced*, arranged in alphabetical order and cited by numbers or dates. In citing papers with two or more authors, only the first author should have the initials after the surname. The address of the author should appear at the end of the text, before the bibliography.

Each article will be restricted to two pages of half-tone illustrations or three of zinc line-engravings for each article, the total cost not to exceed $25. If figures are mounted on board, the cost of routing may be such as to restrict the space allowance substantially. Should the cost of cuts exceed $25, the author will be asked to pay the excess.

To comply with the International Rules, it is recommended that contributors furnish *brief* Latin diagnoses of all new species and genera when their manuscript is submitted for publication.

PRICE LIST OF REPRINTS

	4pp. 1 to 4	8pp. 5 to 8	12pp. 9 to 12	16pp. 13 to 16	20pp. 17 to 20	24pp. 21 to 24	28pp. 25 to 28	32pp. 29 to 32
50 Copies	$4.40	$7.00	$11.00	$11.45	$15.00	$17.15	$19.80	$21.15
100 Copies	5.25	8.35	12.75	14.10	18.00	21.15	24.65	26.45
Additional Copies per C..	1.75	2.65	4.40	5.30	6.00	8.00	9.70	10.60

For 500 copies deduct 5%; for 1000 copies or more deduct 10%.

Covers: For first 50 covers, $4.80; additional $3.45 per C.

For more than 32 pages add cost per schedule to make total. Example: for 44 pages add cost for 32 pages and 12 pages.

Note: For any reprints requiring additional composition or changes, either in text or cover, an extra charge will be made.

LANCASTER PRESS, INC.

LANCASTER, PA.

Partial List of Publications of The New York Botanical Garden

Mycologia, bimonthly; devoted to fungi, including lichens; containing technical articles and news and notes of general interest. $8.50 a year.

Established by The New York Botanical Garden in 1909, in continuation of the Journal of Mycology, founded by W. A. Kellerman, J. B. Ellis, and B. M. Everhart in 1885. Edited by William Alphonso Murrill, 1909–1924. Edited by Fred Jay Seaver, 1924–1946; by Alexander H. Smith, 1946–1950. Edited by G. W. Martin, 1951–1957. Beginning with January, 1958, the official organ of the Mycological Society of America.

North American Flora. Descriptions of the wild plants of North America, including Greenland, the West Indies, and Central America. Planned to be completed in 34 volumes. Roy. 8vo. Each volume to consist of four or more parts. [Not offered in exchange.] Volumes 1–10 devoted to fungi.

Vol. 1, part 1, 1949. Myxomycetes.

Vol. 2, part 1, 1937. Blastocladiaceae, Monoblepharidaceae, Saprolegniaceae, Ectrogellaceae, Leptomitaceae.

Vol. 3, part 1, 1910. Nectriaceae-Fimetariaceae.

Vol. 6, part 1, 1922. Phyllostictaceae (pars).

Vol. 7 (now complete), parts 1–15, 1908–1940. Ustilaginaceae-Accidiaceae.

Vol. 9 (now complete), parts 1–7, 1907–1916. Polyporaceae-Agaricaceae (pars).

Vol. 10, part 1, 1914; parts 2 and 3, 1917; part 4, 1924; part 5, 1932. Agaricaceae (pars).

Series II, part 1, 1934. Tuberales.

Brittonia, quarterly; beginning in 1957, official publication of the American Society for Plant Taxonomists. $7.50 a year.

Economic Botany, quarterly; review and research articles dealing with plants useful to man; established 1947; beginning 1959, the official journal for The Society for Economic Botany. $8.00 a year.

The Memoirs of The New York Botanical Garden, issued irregularly beginning in 1900; approximately 500 pages per volume. $10.00 a volume.

The Myxotrons of North America. By Robert Hagelstein. 1944. $7.50.

NOTICE

Inquiries in regard to subscriptions to the above journals should be addressed to The New York Botanical Garden.

Inquiries in regard to all back numbers and reprints of back numbers of the above journals, and the book, should be addressed to Stechert-Hafner, Inc., 31 East 10th Street, New York 3, New York.

The Garden Journal of The New York Botanical Garden, bimonthly, illustrated, containing news, book reviews, and non-technical articles on botany, exploration, and horticulture. Free to all members of the Garden. To others, 55 cents a copy, $3.00 a year domestic, $3.50 foreign. A continuation of the Journal of The New York Botanical Garden, fifty-one volumes.

The New Britton and Brown Illustrated Flora of the Northeastern United States and Adjacent Canada. By Henry A. Gleason. 3 volumes. 1952, slightly revised 1958. List price $35.00 per set; shipping charge $1.00. Succeeds the Illustrated Flora by Nathaniel L. Britton and Addison Brown. Includes descriptions and drawings of the plant species, from ferns to orchids, which grow without cultivation in the area extending from the St. Lawrence River to Virginia and westward to Missouri and Minnesota.

NEW YORK BOTANICAL GARDEN
Bronx Park, New York 58, N. Y.